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"ETIOLOGY OF LEAF SPOT DISEASES OF IVY GOURD (*Coccinia grandis* (L.) Voigt) AND THEIR MANAGEMENT"

By

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THESIS

Submitted in partial fulfilment of the requirement for the degree of

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Faculty of Agriculture
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Department of Plant Pathology

COLLEGE OF HORTICULTURE

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2003

DECLARATION

I hereby declare that the thesis entitled **"Etiology of leaf spot diseases of ivy gourd (*Coccinia grandis* (L.)Voigt) and their management"** is a bonafide record of research work done by me during the course of research and the thesis has not previously formed the basis for the award to me if any degree, diploma, associateship, fellowship or other similar title, of any other university or society.

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CERTIFICATE

Certified that the thesis entitled **"Etiology of leaf spot diseases of ivy gourd (*Coccinia grandis*(L.)Voigt) and their management"** is a record of research work done independently by **Ms Deepa Davis C.** under my guidance and supervision and that it has not previously formed the basis for the award of any degree, diploma, fellowship or associateship to her.

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Deepa Davis. C.

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Introduction

1. INTRODUCTION

Cucurbitaceous vegetables, consisting of thirty species of cultivated plants, possess an important role in human diet. Ivy gourd (*Coccinia grandis* (L.) Voigt) an important cucurbitaceous vegetable, grown for it's edible fruits, which are consumed either fresh or after cooking. It is grown in Africa,SouthEastern Asia, Latin America, Australia and other tropical countries. In India, it is cultivated mainly in Kerala and parts of Western India.

Ivy gourd is a climbing or prostrate perennial herb, growing throughout India, posses glabrous stem, long tuberous roots and ovoid or elliptic fruits. Flowers are white and the fleshy fruits appear as green with white stripes, when young, turning scarlet at maturity. Fruits are rich source of vitamin A and C and minerals like iron and calcium. They are used as vegetable when green, eaten either fresh when ripe or cooked. It has got therapeutic value as per ayurvedic system of medicine particularly in the treatment of diabetes, bronchitis and skin diseases. It is an under exploited vegetable, the cultivation of which has gained importance in recent years. Released varieties are not available in Kerala and the farmers grow their own material.

Recently, it is noticed that this plant is infected by leaf spot diseases which cause severe damage to leaves, and thereby reducing the vigour and yield of the plant. Infection was also seen on fruits, which reduces its market value. But the crop had not received much attention by the researchers in Kerala.

Munjal *et al.* (1959) reported *Cercospora cocciniae* from leaves of ivy gourd, which was collected from Punjab. Rao (1964) recorded the anthracnose disease on fruits of ivy gourd caused by *Colletotrichum lagenarium* from Pune. Laxminarayana and Reddy (1976) explained the symptoms produced on fruits of ivy gourd by *Colletotrichum gloeosporioides*.

The present study is aimed to find out the etiology of leaf spot diseases of ivy gourd occuring in Kerala as well as to evolve an effective management practice. There were reports on chemical control of leaf spot and fruit diseases in cucurbits. As its leaves and fruits are consumed as green especially by the diabetic patients, the disease management using chemicals is not advisable. Use of biocontrol agents and plant extracts are an alternative for this. In this context, the study entitled "Etiology of leaf spot diseases of Ivy gourd (*Coccinia grandis* (L.) Voigt) and their management" was taken up with the following objectives,

1) To study the etiology of leaf spot diseases of ivy gourd
2) To find out the seasonal influence on the incidence of leaf spot diseases
3) Screening for host resistance of ivy gourd genotypes against leaf spot diseases
4) Management of leaf spot diseases by using fungicides, botanicals and antagonistic microorganisms.

Review of Literature

2. REVIEW OF LITERATURE

Ivy gourd is an important cucurbitaceous vegetable grown in almost all parts of India. The plant is getting popularity day by day due to its profitable proposition and also due to its nutritive value and medicinal property. Even though this plant is hardy, several diseases especially leaf and fruit spots are noticed in many vegetable fields where it is cultivated in large scale. Many diseases of ivy gourd were reported from different parts of India, but a perusal of literature revealed no reports from Kerala.

2.1 Symptomatology

Jain (1950) first reported a new leaf spot disease in *Cephalandra indica Naud* (*Coccinia indica Wt.&Arn.*) caused by *Cercospora* species. He also described the symptoms of the leaf spot which started as minute yellow dots on upper side and turned brown, and then to whitish from centre outwards with a brown ring. The matured spots were measuring 0.5 to 4 mm in diameter with depressed white centre where cluster of conidiophores formed which appeared as black dots. The infected leaves turned yellow and dried off.

Rao (1962) described the symptoms produced by *Cercospora citrullina* on different cucurbitaceous host plants. He reported that the infection spots were oval to circular in shape and measured 2-8 mm in diameter.

Symptoms developed on fruits of ivy gourd due to the infection of *Colletotrichum lagenarium* were described by Rao (1964). He reported that the infection appeared as minute dark-brown, soft, oval to angular, slightly sunken spots over green fruits.

Laxminarayana and Reddy (1976) explained the symptoms produced by *Colletotrichum gloeosporioides* on fruits of ivy gourd as water soaked areas with tissue disintegration and depression in the centre of the spots with light brown margin. They were also noticed acervuli formation in the pinkish white centre of spots during the advanced stage of infection.

In the study of Alternaria blight of cucurbits, Bharghava and Singh (1985) described the symptoms produced on watermelon. They reported that the symptoms started with yellow or brown spots with bright yellow halo and later it became larger with frequent zonations and marginal necrosis.

A severe leaf spot disease of cucumber caused by *Alternaria alternata* was first observed by Vakalounakis and Malathrakis (1988). They observed that the lesions ranged from the size of pinprick to 5 cm in diameter, with necrotic tissue on most of their area and surrounding a yellow zone.

Khan (1989) reported that the symptoms of powdery mildew of cucurbits appear first on lower surface and gradually spread to the upper surface.

Bhartiya *et al.* (2000) described three new species of *Cercospora* associated with vegetable crop viz. chilly, tomato and brinjal. They observed a spreading type of symptom on chilly showing necrotic greyish white centre with brown margin and irregular shape where as in tomato, leaf spots were smaller in size, circular to irregular, necrotic with white centre and dark brown margin. Symptoms of brinjal leaf spot was similar to this, but the spots were dark brown centered with brown margin.

2.2 Etiology of leaf spot diseases

A perusal of literature revealed no reports of *Cercospora* sp. on *Coccinia indica* from Kerala. Jain (1950) first reported an unidentified species of *Cercospora* in *Cephalandra indica* (*Coccinia indica* Wt.&Arn.) from North India. Chiddarwar (1959) presented a paper which included details of different species of *Cercosporae* of the Bombay State. Among the new species, he reported *C. momordica* on the leaves of *Coccinia indica*.

Munjal *et al.* (1959) described a new species, *Cercospora. Cocciniae*, which was collected from Punjab in 1954. Later, Rangaswami and Chandrasekaran(1961) reported *C. cocciniae* on the leaves of ivy gourd and confirmed the identity of their isolate with *C. cocciniae* described by Munjal *et al.* in 1959.

Rao (1962) reported *C. cocciniae* on *C. indica* from Hyderabad. Again Rao (1964), first reported *C. lagenarium* on ivy gourd fruits.

A report on Alternaria blight of watermelon causing serious losses through out Rajasthan was given by Khandelwal and Prasada (1970). They also observed the infection on muskmelon and cucumber by the same fungus in their host range study. Hasan *et al.* (1974) reported the incidence of *C. lagenarium* on ivy gourd fruits.Laxminarayana and Reddy (1976) isolated *C. gloeosporioides* from spots developed on fruits of *Coccinia indica* from Andra Pradesh.

In Karnataka, *C. lagenarium* causing anthracnose was newly recorded on watermelon by Hiremath (1981). Fruit rot of pointed gourd caused by *A.alternata* and *Glomerella cingulata* was described by Kritagyan and Singh (1980).

Naqvi (1985) reported fruit rot of *C. indica* caused by *Colletotrichum destructivum*. Due to this infection, considerable losses were reported under field condition, and infection occurred at all stages of fruit growth. A new leaf blight and fruit rot of watermelon caused by *Alternaria alternata* was reported by Narain *et al.* (1985).

A severe leaf spot disease of cucumber caused by *A. alternata* was reported by Vakalounakis and Malathrakis (1988) from Greece. Bedi *et al.* (1993) isolated and reported *A. alternata* from the infected leaves, tendrils and stems of watermelon in Punjab and pathogenicity was confirmed.

Rego *et al.* (1995) reported and studied the host range of *C.orbiculare* in cucurbitaceous hosts. Some new Cercospora species associated with solanaceous crops in North India was reported and described by Bhartiya *et al.* (2000).

2.3 Morphology of causal organisms

The morphological characters of newly identified *Cercospora* species causing leaf spot on *Coccinia* were studied in detail by Jain (1950). On examination of infected host tissue, he found that the hyphae of the pathogen was mostly intracellular, hyaline to olivaceous brown in colour strongly septate and frequently branched. The conidiophores were brown, 0 to 5 septate and 1 to 5 times geniculate and 12 to 120 µm x 3 to 6 µm in measurement. The conidia were hyaline to olivaceous, filiform to obclavate, tapering towards the free end, 0 to 12 septate and measured 21.54 to 141.6 µm x 2.34 to 6 µm.

Rao (1962) described the characters of *C. citrullina*. He reported that the conidiophores of this species were found as groups of 5-25, dark brown, 4-5 septate, rounded at the apex, unbranched, straight or curved, geniculate, measuring

54-144 µm x 3.6-8.0 µm. Conidia, hyaline, straight or curved, slender, long, 5-20 septate and measured 72-288 x 3.6-6.4 µm.

According to Rao (1964), conidia of *C. lagenarium* were hyaline, subcylindrical with rounded ends, one celled, few having central oil globules, oblong to ovate-oblong and measures 14.7-30 µm x 5.2-6.5 µm. Acervuli dirty brown to dull pink, loose and setae few short, slightly blunt at the apex and dark brown.

Bharghava and Singh (1985) noted the characters of pathogen causing Alternaria leaf blight of cucurbits. Spore length ranged between 36-108 µm, beak length 36-41 µm having 1-4 cross septa and 1-3 longitudinal septa.

2.4 Cultural characters of causal organisms

As stated by Jain (1950), leaf portions showing symptoms of *Cercospora* leaf spot were inoculated in rice meal agar plates and the fungus started growth on the third day of inoculation. He observed dark green coloured colonies of the fungus with white fluffy aerial mycelium in the centre. The aerial mycelium looked greyish white, hyphae showed profuse branching and anastomosis, had a width of 0.78 to 4.5 µm. He also observed conidia in the culture which were sub hyaline, and cylindrical on the tips of conidiophores and they were brown, multi-septate and geniculate with scars of spore attachment.

Carrot leaf extract agar has been reported to be good for the growth and sporulation of some species of *Cercospora* (Kilpatrick and Johnson, 1956). The optimum pH for their growth was between 6 and 7 and the temperature around 25°C. They tested different medium for spore production, but observed spore production only in agar medium prepared from carrot leaves steamed without pressure for 1 hour. The spores were found only at the edge of the colonies before the fungus had developed a heavy growth of mycelium. They reported that substance in carrot leaves that induces sporulation was changed chemically by pressure sterilization.

Among different media tested for *Cercospora* species occurring on cucurbitaceous hosts in South India by Rangaswami and Chandrasekaran (1962) found carrot leaf extract agar and carrot agar as best for isolation. The resultant colonies of *C. citrullina* were compact and raised, dark grey to black with effuse white mycelium in the central region. Reverse of the colonies were dark green, some sporulation were also noted in carrot leaf extract agar medium.

Bhargava and Singh (1985) observed the cultural characters of *Alternaria cucumerina* isolated from cucurbits in malt extract medium and reported that the colonies were circular, cottony, smooth, olivaceous green with definite concentric zonations.

Khandar *et al.* (1985) found that sporulation of *Cercospora canescens* causing mung bean leaf spot was good in carrot leaf juice-oat meal agar. They concluded that glucose and sucrose supported the growth of fungus at a temperature of 25°C.

2.5 Spore germination and histopathological studies

Hashioka (1937) reported conidia of cucurbit powdery mildew organism germinate from 22° to 31°C with a peak at 28°C.Jain (1950) found that at room temperature conidia of Cercospora species germinate within 12 to 15 hours, usually from the terminal cell and occasionally from the intermediate cell. Anastomosis between germ tubes and germinating conidia was observed occasionally.

Brown (1975) found that the spore of *C.gloeosporioides* affecting citrus plants germinated on the surface of citrus plants on the surface of citrus fruit and formed appressoria which in turn produced infection hyphae. Tebust *et al.* (1978) observed that spore of *C.gloeosporioides* infecting Aeschynomene germinated and produced appressoria in 4-5 hours.

Percent spore germination of *Cercospora canescens* infecting mung bean were calculated by Khandar *et al.* (1985). They noted the optimum conidial germination after 6 hours of incubation at a temperature of 27°C and at pH 6 and 7.

Raghuram and Mallaiah (1996) studied the conidial germination of Cercospora species infecting legumes, by slide germination method at laboratory temperature of 28-32°C. They noted that the basal cell germinated first followed by apical and intercalary cells.

Munjal and Gupta (1965) observed during the study of host parasite relations of the anthracnose of *Celosia* that the cells of diseased tissue lost their shape and later the hyphae collected underneath the epidermis from stroma, from which conidiophores were produced. Tebust *et al.* (1978) reported that when the weed plant *Aeschynomene* was infected with *C.gloeosporioides* the mycelium grew within the cortex, cambium, xylem and pithway tissues and death of seedlings occurred.

2.6 Effect of toxin on disease development

Toxins were produced by many fungi causing leaf spot diseases. Goodman (1960) reported that toxin produced by *Colletotrichum fuscum* induced spots on tomato foliage and affects plants, which were not directly attacked by the pathogen.

Sharma and Sharma (1969) reported that production of toxin by *C. gloeosporioides* induced die-back in citrus. Nair and Ramakrishnan (1973) conducted studies on toxin production by *C. capsici* causing leaf spot in turmeric. When leaves were treated with exo and endotoxin of pathogen, symptoms were noticed within four hours of inoculation.

Exo and endotoxins of *C. gloeosporioides* causing leaf spot of *Plumbago indica* was studied by Varma (1991) and observed that endotoxic metabolites produced symptoms much earlier than exotoxic metabolites.

2.7 Host Resistance

Cultivating resistant varieties is the most effective and economical means of controlling plant diseases and getting good yield. Williams (1977) screened 5000 cowpea lines against important bacterial and fungal diseases including *C. lindemuthianum* under field conditions and found that 16 cowpea lines showed multiple resistance to the diseases. Sohi and Rawal (1983) screened 141 cowpea varieties against *C. lindemuthianum* and found 21 varieties as resistant to the pathogen.

Kabitarani and Bhagirath (1991) conducted a study to find out resistant lines of 12 *Cucurbita moschata* germplasm against powdery mildew under field conditions. None of the germplasm were immune, but some varieties showed high degree of resistance while others were highly susceptible.

Lebeda (1991) studied six muskmelon lines for resistance to eight isolates of cucurbit downy mildew originating from cucumbers. He found that only one line was highly resistant and it was also resistant to powdery mildew under green house and laboratory conditions.

Pans and More (1995) screened melons for resistance to downy mildew and reported that wild cucumber species, *C. figarei* exhibited high level of resistance. Raju and Peter (1995) reported CG1, CG2, CG4, CG9, CG23, CG27 and CG37 as promising genotypes in ivy gourd considering the characters like fruit length, fruit weight, and fruit girth.

2.8 Seasonal occurrence

Mordue (1971) reported that a temperature of 15-26°C and humidity greater than 92 per cent favoured symptom expression of bean anthracnose caused by *C. lindemuthianum.*

Singh (1985) pointed out that germination and growth of the powdery mildew fingers in cucurbits occurred best at 22° to 27°C. The fungus occurred in epidemic form only when there was more than average rainfall. Disease development can occur at 20° to 30°C with optimum at 25°C under conditions of 100 per cent relative humidity for at least 18 hours.

Ullasa and Amin (1988) showed that day temperature of 25-30°C and relative humidity 75 per cent favoured the injection of *Luffa acutangula* by *Pseudoperenospora cubensis* causing downy mildew.

During the study of seasonal occurrence of fungal diseases in medicinal plants, Varma (1991) reported that *Colletotrichum* infection was seen during January to May and *Alternaria* and *Cercospora* were predominant during the period May-June to October-November, synchronizing with two monsoon seasons.

Thakur and Khare (1992) reported that *Vigna radiata* was severely affected by *C.dematium* and *C.lindemuthianum* during monsoon season. Here, disease intensity was positively correlated with RH and rainfall and negatively correlated with maximum and minimum temperature. Thakur and Khare (1993) observed that moderate temperature of 25^{0}C and RH 90-100 per cent were highly favourable for germination and sporulation of *C.dematium* and *C.lindemuthianum* on *V. radiata.*

Gandhi *et al.* (1997) determined the functional relationship between bottle gourd leaf spot caused by *C. orbiculare* and meteorological factors. They found that the apparent rate was maximum when maximum temperature was 32°C, minimum temperature 25°C and relative humidity was more than 90 per cent in the morning and 80 per cent in the evening combined with rainfall.

Kumar (1999) observed maximum infection on cowpea by the pathogen *C.lindemuthianum,* and yield loss during the southwest monsoon season.

2.9 Disease management

Efforts to control the leaf spot pathogens should be started from very early stage of disease, immediately after the expression of symptom. Now a days, the effect of plant protection chemicals, botanicals and antagonists in the management of disease has practical importance. So many workers have conducted studies on these aspects.

2.9.1 Fungicides for management of leaf spot diseases

Several workers studied the effect of plant protection chemicals against leaf spot pathogens. A comparative study on toxicity and biological spectrum of three copper fungicides and two organic fungicides and their effect on *Alternaria tenuis Auct.* were done by Misra and Singh (1965). They tried Bordeaux mixture, Blitox-50 and Fytolan and two organic fungicides viz. Dithane Z-78 and Captan to assess their relative merits. Captan inhibited the pathogen to the most, followed by Dithane Z-78 and Bordeaux mixture. Blitox and Fytolan were less effective compared to others.

Misra and Singh (1971) tested the efficacy of Bordeaux mixture, Blitox-50 and Fytolan and organic fungicides, Dithane Z-78 and Captan against the growth of *A.tenuis* and *Helminthosporium oryzae* Breda de Haan by poisoned food technique. They recorded Captan as superior to all the other fungicides for both the organisms followed by three copper fungicides. Higher concentration of Dithane Z-78 was required to produce 50 per cent inhibition and thus ranked last.

During the control trials against false mildew by *Pseudoperenospora cubensis* (B.&C.) Rostow causing damage on cucumber, Akalycin and Guncu

(1978) tried Antracol (propineb) and Dithane M-45 (Mancozeb). At two per cent concentration both gave 94.7 per cent and 95.6 per cent control respectively.

Chauhan *et al.* (1980) tested the efficacy of some systemic and nonsystemic fungitoxicants for the control of anthracnose caused by *C.lagenarium* in bottlegourd. In field spraying, the best control was achieved with Difolatan (Captafol) followed by Bavistin and Benomyl. In the chemical control of *C. lagenarium* of bottle gourd, Madaan and Grover (1980) found that spraying with Difolatan (captafol) or Blitox (Copper oxy chloride) gave the best control of leaf anthracnose and fruit scab. Here Benomyl and Thiophanate methyl were less effective compared to others.

In watermelon, chemical and cultural control against *C.lagenarium* were done by Peregrine and Ahmad (1983). From the field trials, they recommended Benomyl and Mancozeb or Captafol for control and they also suggested a high standard of management during August to December, the wettest months of the year.

Suhag *et al.* (1983) conducted an experiment to test the efficacy and economics of different fungicides for the control of leaf spot on bottle gourd caused by *Cercospora* sps. It was observed that among the five fungicides tried Bavistin (0.1 per cent) gave more than 80 per cent disease control and Dithane M-45 (0.2 per cent) provided maximum monetary benefit. Difolatan was the next best giving about 78 per cent disease control which was better than Dithane M-45 in controlling disease. Blitox (0.2 per cent) provided 75 per cent reduction in disease over the control.

Peregrine *et al.* (1984) found that Benlate (Benomyl) and Dithane M-45 (Mancozeb) performed best in the field tests conducted against *C. lagenarium* infection in watermelon. They also observed the effectiveness of the same fungicidal treatments to control *C. citrullina* and *Didymella bryoniae* causing minor diseases in the crop.

When 11 fungicides were tested against chilli fruitrot caused by *Colletotrichum capsici* and *Alternaria solani*, Foltaf (0.2 per cent captafol) gave the most effective control followed by Fytolan (0.25 per cent) and Bavistin (0.1 per cent) (Jayasekhar *et al.*, 1987). They found that treatment with Fytolan resulted in maximum yield followed by Foltaf and Bavistin.

Bhardwaj (1991) found that the sequential application of Captafol, Mancozeb and Copper oxychloride (all at 0.25 per cent), increased yield by 50.5 per cent by reducing the incidence of *A. solani, A. alternata* and other pathogens causing foliar and fruit diseases in tomato.

Bhardwaj and Thakur (1991) obtained the best possible control of leaf spot diseases of *Vigna mungo* caused by *Cercospora cruenta, C. canescens, Colletotrichum truncatum* and *Ascochyta phaseolarum* using sequential application of 0.25 per cent mancozeb, 0.1 per cent Carbendazim and 0.25 per cent Captafol.They also observed 34.6 per cent increase in yield due to this application.

During the evaluation of some systemic and nonsystemic fungicides against *C. lagenarium* in watermelon, Hiremath (1991) found that the application of Captafol followed by Carbendazim and Benomyl reduced the disease and increased the yield by 55.6-87.5 per cent. To get control over the disease, he recommended two applications of Captafol (0.03 per cent) at 20 days interval.

Tahir *et al.* (1991) tested 7 fungicides in field against *Alternaria porri* in onion causing purple blotch disease. Here treatments with Cupravit (Copper oxychloride), Ridomil MZ-72 and Penncozeb (Mancozeb) reduced the disease as well as increased bulb yield of the crop.

Also, in field evaluation of fungicides against potato early blight caused by *A. solani*, Guddewar *et al.* (1992) found that the best control and highest yield were obtained with Mancozeb(2 kg/ha), Captan(2.5 kg/ha) and Copper oxychloride (2.5 kg/ha). Fungicides were applied 53 days after planting with 3 sprays at an interval of 7 days.

In the chemical control trials by Maheshwari and Gupta (1992) against *C. lagenarium* in bottlegourd, Dithane M-45 (Mancozeb) followed by Blitox-50 (Copper oxychloride) were found most effective. Petkar and Rai (1992) conducted a study on the effect of fungicides on activity and growth of *A.alternata*. They concluded that Captan, Cosan, Thiram, Zineb and Sandofix inhibited the polygalactouronase and cellulase activity and the secretions by *A. alternata* and also these fungicides inhibited the growth of the pathogen.

Bedi *et al.* (1993) tested the effectiveness of fungicides against *A. alternata* in watermelon. In lab tests, Rovral and Dithane M-45 (Mancozeb) were found effective against the pathogen. Yadav and Narain (1993) studied the chemical control of Alternaria blight of chickpea caused by *A.alternata*. Here, among 10 different fungicides tested, the best control was given by Difolatan (Captafol) at 0.2 per cent, applied 3 times at 10 days interval. San619F (0.1 per cent), Dithane M-45 (Mancozeb) and Cuma L (Ziram) were less effective.

Kumar (1999) found that among different fungicides tried, Mancozeb was the most effective one to control the anthracnose in cowpea caused by *Colletotrichum lindemuthianum*.

2.9.2 Botanicals in disease control

Continuous use of fungicides in controlling diseases is not advisable due to the cost and adverse environmental hazards, besides development of resistance against pathogen. So the utilization of naturally occurring substances in plants with fungicidal properties is an effective method to overcome such problems.

Lakshmanan (1990) tested 10 plant extracts against cotton ball rot pathogen (*Cornespora cassiicola*) and found that garlic was most effective, inhibiting mycelial growth by 95.8 per cent. Fungicidal properties of neem and blue gum against *Sclerotium rolfsii* was tested by Singh and Dwivedi (1990) and found that neem oil was most effective against the pathogen.

During the study of powdery mildew on cucumber plants caused by *Erysiphae cichoracearum*, preventive treatments including 1-5 per cent garlic extract, 0.2 to 1 per cent *Equisetum arvense* extract, 3 per cent liquid soap and horse manure extract were tested by Qvarnstrom (1992). He found that 5 per cent garlic extract was most effective in preventing the disease and all other treatments were less effective compared to garlic.

Ahmad and Prasad (1995) found that the conidial germination of *Fusarium scirpi* and *Cochliobolus spicifer* effecting sponge gourd fruits were reduced by treating them with extracts of *Azadirachta indica, Catharanthus roseus, Datura fistulosa, Lantana camara* and *Ocimum sanctum*. Barros *et al.* (1995) reported the inhibition of *A. alternata* and *Alternaria longipes* with bulb extracts of garlic, against mycelial growth.

Sinha and Saxena (1995) found that phytoextracts of *A. indica, L. camara, O.sanctum* and *D. fistulosa*, almost fully inhibited the spread of Fusarium rot of sponge gourd. Antifungal properties in these plant extracts were observed against growth of *Alternaria brassicae, A. alternata, C capsici, Fusarium oxysporum* etc.

Simon (1996) observed the effect of *O. sanctum* (10 per cent) in controlling the downy mildew disease in bittergourd. Asha *et al.* (1997) studied in fungitoxic property of ethanol extract of *Ocimum sanctum* on *C. capsici* and found that it was highly effective in controlling the pathogen. Experiment done by Ilyas *et al.* (1997) revealed that neem product, Replin completely inhibited the vegetative growth of *Macrophomina phaseolina* at 0.8 per cent level and it also reduced the sclerotium formation.

Singh and Prithiviraj (1997) reported that Neamzal, a product of neem significantly retarded the growth of *Trysiphae pisi* in *Pisum sativum*. Antifungal activities of extracts of *A. indica, O. sanctum, Allium cepa* and *Allium sativum* have been reported by Ram (1997) and Shivpuri *et al.* (1997). Kurucheve and Padmavathi (1997) assayed five products for fungi toxicity against *Pythium aphanidermatum* causing damping off of chillies and found that extracts of *Allium sativum* bulbs (10%) recorded the minimum mycelial growth followed by *Lawsonia inermis*.

Singh and Majumdar (2001) tested the effect of extracts of some medicinal plants against *Alternaria alternata* causing fruit rot of pomegranate. They found that among the botanicals used maximum inhibition of growth was recorded at 20 per cent concentration for garlic extract followed by turmeric, ginger, datura and neem.

2.9.3 Antagonists in disease control

The genus *Trichoderma* has been demonstrated to be a potential biocontrol agent against plant pathogenic fungi (Liu and Baker, 1980). Biological control, using microorganism against the plant pathogens is an effective and alternative tool in managing the disease and is gaining importance in recent years. Biological control against fungal pathogens has gained considerable attention and appears to be promising as a viable supplement or alternative to chemical control (Natarajan and Manibhushanrao, 1996).

Sychev and Shaprshnik (1982) studied the antagonistic effects of *Trichoderma viride* in relation to some pathogens of *Cucumis sativum* L. They found that *T. viride* inhibited the growth of *Rhizoctonia solani, Sclerotinia sclerotiorum* and many more fungi isolated from cucumber.

When *Penicillium stipitatum* and *Trichoderma harzianum* were used against cucumber damping off caused by *Pseudoperenospora cubensis*, Sharif *et al.* (1988) obtained control over the disease which were equivalent to that obtained with Ridomil (metalaxyl). Das and Nair (1990) reported that application of *T. viride* in soil reduced the intensity of sheath blight disease in rice. They observed that *Chaetomium globosum* and *Fusarium solani* were also effective but not as much as *T. viride.*

Anatgonistic activity of different *Trichoderma* species against *Colletotrichum graminicola* infecting sorghum leaves were studied by Michereff *et al.* (1993). In their study, inhibition zones were observed mainly for *T. viride* and T. *harzianum*. All species could inhibit the growth of pathogen, but alterations in the morphology of pathogen was seen in the case of *T. viride* and *T. pseudokoningii.*

Bankole and Adebanjo (1996) studied the biocontrol of brown blotch of cowpea caused by *C. truncatum* with *T. viride*. Seed dip and soil drenching of this biocontrol agent was found to be very effective. Foliar application also significantly reduced brown blotch incidence in the field.

Menon (1996) reported the *in vitro* inhibitory effect of *T. viride* against the damping off pathogens of solanaceous host.In pot culture experiment, reduction in sheath blight infection in rice was observed when *T. viride* was used for seed treatment(Das *et al.*,1998). Kumar (1999) observed the effectiveness of *T. viride* against anthracnose disease of vegetable cowpea caused by *C. lindemuthianum*

Smitha (2000) reported that among the different isolates of *Trichoderma*, *T. longibranchiatum* was superior in inhibiting the pathogen, *Rhizoctonia solani* causing foliage diseases of leafy vegetable amaranthus. *T. viride, T. longibrachiatum* and *T. harzianum* were found parasitic on *Phytophthora capsici* effecting black pepper as evidenced by mechanisms like coiling, penetration and disintegration of the hyphae (Vijayaraghavan, 2003).

P. fluorescens produced siderophores which was antagonistic to *R. solani* in rice under *in vitro* condition (Sakthivel *et al.*, 1986). Anuratha and

Gnanamanickam (1990) observed that strain pfcp of *P.fluorescens* was effective in controlling bacterial wilt of banana, eggplant and tomato under greenhouse and field conditions.

Thara and Gnanamanickam (1990) reported that *P. fluorescens* and *P. putida* can effectively suppress *Rhizoctonia solani* causing sheath blight in rice. *Pseudomonas* sps. isolated from healthy leaves of cluster beans was found inhibitory to the growth of pathogen, *Xanthomonas campestris* Pv. *Cyamopsodis* (Saini and Parashar, 1991).

Casida and Lukezic (1992) reported the reduction in severity of leaf spot diseases of alfalfa and tomato caused by *Alternaria solani, Pleospora tarda* etc. using the application of *Pseudomonas* strain 679.2. They also found that the treatment produced no adverse effects on plants.

Rabindran and Vidhyasekaran (1996) recommended the use of *P. fluorescens* at different stages including seed treatment, root treatment, soil application and foliar spraying for the control of sheath blight in rice. Kamala *et al.* (1998) reported that combination of methods of application viz., seed treatment, seedling root tip and foliar spray of talc based formulation containing *P. fluorescens* gave 60-70 per cent control of sheath blight in rice.

Srinivasan and Gunasekaran (1998) reported the reduction of incidence of leaf spot disease of coconut palms using the antagonist *Pseudomonas fluorescens*. Antagonist can inhibit the growth of pathogen, *Colletotrichum gloeosporioides* to considerable level. Among different antagonists tested against bacterial wilt in solanaceous vegetables, *T. viride, T. harzianum, T. pseudokoningii, A. niger, Bacillus subtilis* and *P. fluorescens* were found most effective against *Ralstonia solanacearum* (Manimala, 2003).

2.10 Disease intensity and severity

Khandelwal and Prasada (1970) found that *A. cucumerina* causes serious losses on watermelon throughout the year. Disease intensity varied from 2-63 per cent giving a net loss in yield from 7-54 per cent. They noted that fungus infected muskmelon, round gourd, pumpkin and cucumber besides watermelon.

Bhargava and Singh (1985) reported the severity of Alternaria blight on cucurbits as 12-80.7 per cent. They observed maximum yield losses of 88.3 and 80 per cent on watermelon and pumpkin respectively.

In the case of Colletotrichum fruit rot of *C. indica*, considerable losses in yield were reported by Naqvi (1985). Infection occurs at all stages of fruit growth and 60-75 per cent disease incidence was recorded. Severe disease intensity and losses in solanaceous crops like chilly, tomato and brinjal due to the infection by Cercospora species were reported by Bhartiya *et al.* (2000).

Materials and Methods

3. MATERIALS AND METHODS

The present study on 'Etiology of leaf spot diseases of Ivy gourd (*Coccinia grandis* (L.)Voigt) and their management' was conducted in the Department of Plant Pathology, College of Horticulture, Vellanikkara, Thrissur during the period from January 2002 to August 2003. The details of the materials used and the techniques adopted for the investigation are described below.

3.1 Survey and collection of diseased samples

A survey was conducted in the vegetable field of Department of Olericulture, College of Horticulture, Vellanikkara as well as in the farmer's field at Cherumkuzhi and Elanad to study the occurrence of leaf spot diseases of ivygourd and to collect diseased samples for the experimental studies.

3.2 Isolation of pathogens associated with leaf spot diseases

The leaves showing different types of leaf spot symptoms were collected separately from the above mentioned areas and brought to the laboratory and were observed under the microscope by preparing the slides from infected areas. Samples were then washed under tap water and dried using blotting paper. Small bits of infected leaf portions along with some healthy areas were surface sterilized with 1 per cent sodium hypochlorite and then washed in three changes of sterile water. Different media like Potato Dextrose Agar (PDA), Carrot Leaf Extract Agar (CLEA) and Coccinia Leaf Extract Agar (CoLEA) were used for the isolation of the different pathogens associated with leaf spot symptoms. The fungi grown on the media were purified, subcultured and maintained for further investigations..

Likewise, the isolation of the pathogen was also done from the infected fruits collected from the three locations. The composition of different media used for isolation is given in Appendix I.

3.3 Pathogenicity

The pathogenicity of the organisms associated with leaf spot and fruit spot diseases were studied by artificial inoculation under *in vitro* and *in vivo* conditions.

3.3.1 *In vitro* condition

Healthy leaves collected from the field, were washed under tap water and then surface sterilized with 70 per cent ethyl alcohol. The leaves were inoculated separately on both surfaces with the mycelial growth of the isolates as well as by spraying spore suspension of pathogen having a concentration of 10^6 spores/ml using an atomiser, with and without giving pinprick. Leaves inoculated with sterile water served as control. The inoculated leaves were kept under humid chamber and observed daily for the symptom appearance. The pathogens were reisolated from the inoculated infected leaves and compared with the original culture.

Fruits were also inoculated with the isolated pathogens and observed for the symptom expression.

3.3.2 *In vivo* condition

Stem cuttings of ivygourd were grown in small earthern pots of 20 cm diameter. When it attained 8-10 leaf stage, five middle leaves were inoculated separately with the different isolates as mentioned above. Plants inoculated with sterile water were kept as control. The inoculated plants were placed under humid chamber and observed for the symptom expression. The pathogens were reisolated from the inoculated plants showing leaf spot symptoms and compared with the original culture.

3.4 Symptomatology

Symptoms produced by different leaf spot pathogens on the leaves and fruits of ivy gourd under natural and artificial conditions were studied in detail.

3.5 Identification of pathogens

The pathogens associated with leaf spot diseases were identified based on the morphological and cultural characters.

3.5.1 Cultural and morphological characters of the pathogen

Cultural characters of the pathogen such as rate of growth, colour and formation of fruiting bodies in the selective media were studied in detail.

Morphological characters of pathogens from the naturally infected plants as well as from the pure culture were studied. Size of hyphae, conidia, conidiophores and fruiting bodies of different pathogens were recorded. Camera lucida drawings and photomicrographs of these organisms were made. These characters were compared with the descriptions of CMI to identify the pathogens.

3.6 Histopathological studies

Thin sections of leaves infected with different leaf spots were stained with saffranin and observed under microscope to study the histopathological changes brought by the different pathogens.

3.7 Studies on spore germination of different pathogens

Spore suspension of the pathogens were prepared in sterile water from the pure culture as well as from the naturally infected leaves. Transferred few drops of the spore solution to a sterilized, clean cavity slides with micropipette and covered with glass cover slip. The slides were observed under the microscope and noted the number of spores in a microscopic field. The slides were then kept inside humid chamber for germination of spores. Observation on number of spores germinated was taken at hourly intervals for 10h.

3.8 Effect of toxin on disease development

An *in vitro* experiment was conducted to evaluate the effect of toxins produced by the pathogens on the development of different leaf spot diseases.

Stem cuttings of ivygourd were raised in small pots, with two plants in each pot. Fifty ml of the selective liquid medium was taken in each 250 ml flask and sterilized by autoclaving. The broth was inoculated with mycelial discs of 8 mm diameter obtained from the actively growing seven day old culture of each fungus. These flasks were incubated at room temperature till the fungal growth fully covered the surface of the broth. After the desired period of incubation, medium containing the fungal mycelium was filtered through sterile Whatman No.1 filter paper to obtain exotoxin. The mycelial mat of each fungus was homogenized separately with two times of water and centrifuged at 1000 rpm for 15 minutes. Pellets were discarded and the supernatent was taken which contain endotoxin. Plants were spotted separately with endotoxin and exotoxin of each fungus and three replications were maintained for each treatment. Cuttings were covered to maintain the humidity and observed daily for the development of symptoms. Plants sprayed with sterile water served as control.

3.9 Reactions of different ivy gourd genotypes to leaf spot diseases

Nineteen ivy gourd genotypes grown in the vegetable fields of Olericulture department, Vellanikkara were screened for disease resistance to leaf spot diseases by recording percent disease incidence and severity.

3.9.1 Assessment of disease incidence

For assessing the disease incidence, number of infected plants and total number of plants in each field were recorded and per cent disease incidence was calculated using the formula.

$$\text{Per cent disease incidence (PDI)} = \frac{\text{Number of infected plants}}{\text{Total number of plants observed}} \times 100$$

3.9.2 Assessment of disease severity

Disease severity from each field was calculated by randomly selecting ten plants. Ten leaves were selected randomly from each plant (three from top, four from middle and three from bottom) and were scored using 0-5 scale as detailed above.

Table 1 Score chart for severity of disease on leaves

Grade	Description
0	No symptoms
1	10% leaf area infected
2	>10 - 25% leaf area infected
3	>25 - 50% leaf area infected
4	>50 - 75% leaf area infected
5	>75% leaf area infected

Per cent disease severity was calculated using the formula suggested by Wheeler (1969).

$$\text{Per cent Disease Severity (PDS)} = \frac{\text{Sum of all numerical ratings}}{\text{Total number of leaves observed x Maximum disease grade}} \times 100$$

From per cent disease incidence and per cent disease severity, Coefficient of Infection (CI value) was calculated using the formula suggested by Datar and Mayee (1981).

$$\text{Coefficient of Infection (CI value)} = \frac{\text{Per cent disease incidence x per cent disease severity}}{100}$$

Based on the CI value , the genotypes were categorised into five groups as shown below.

CI value	Category
0-4	Highly resistant(HR)
4.1-9	Resistant (R)
9.1-19	Moderately resistant (MR)
19.1-39	Moderately susceptible (MS)
39.1-69	Susceptible (S)
69.1-100	Highly susceptible (HS)

3.10 Seasonal influence on the incidence of leaf spot diseases

A survey was conducted in three locations of Thrissur district during June 2002 to May 2003. Ivygourd plants grown in the vegetable field of Dept. of Olericulture,Vellanikkara as well as in farmers' fields at Cherumkuzhi and Elanad areas were surveyed at monthly intervals and disease incidence and severity were recorded. These observations were then correlated with the atmospheric temperature, relative humidity and rainfall. Meteorological data were obtained from Department of Meteorology, College of Horticulture, Vellanikkara to study the seasonal influence on the incidence of different leaf spot diseases of ivygourd.

3.11 Management of leaf spot diseases

The effectiveness of fungicides, botanicals and antagonistic organisms against leaf spot pathogens was tested under laboratory conditions. The fungicides and botanicals were evaluated by Poison Food Technique (Zentmyer, 1955). The antagonistic efficiency was evaluated using dual culture method as suggested by Skidmore and Dickinson (1976).

3.11.1 *In vitro* evaluation of fungicides against leaf spot pathogens

For *in vitro* evaluation 100 ml of selective media was taken in 250 ml conical flask and sterilized at 1.05 $kgcm^{-2}$ pressure for 20 minutes. Required quantity of chemicals were mixed with the media to get desired concentrations and poured into sterilized Petridishes @ 20 ml per plate. Mycelial discs of 8 mm diameter were cut from actively growing seven day old culture of the fungus and placed at the centre of each Petridish containing poisoned medium. Five replications were maintained for each concentration of all fungicides. Media without the fungicide served as control. The inoculated Petridishes were incubated at 28 ± 2^0C. The diameter of the fungal colony was recorded upto and when growth in the control plates were fully covered the medium.

The per cent inhibition of growth over control was calculated by the formula suggested by Vincent (1927).

$$\text{Per cent inhibition of growth} = \frac{C - T}{C} \times 100$$

C = Growth of pathogen in control (mm)

T = Growth of pathogen in treatment (mm)

The following fungicides given in Table 2 were used for *in vitro* evaluation.

Table 2 Fungicides used for *in vitro* evaluation against leaf spot pathogens

Sl.No.	Chemical Name	Trade Name	Concentration (Per cent)
1	Copper oxychloride	Fytolan (50WP)	0.1, 0.2, 0.3
2	Copper hydroxide	Kocide101 (77WP)	0.1, 0.2, 0.3
3	Mancozeb	Indofil M-45(75WP)	0.1, 0.2, 0.3
4	Captan	Captaf (50WP)	0.1, 0.2, 0.3

3.11.2 *In vitro* evaluation of selected botanicals against leaf spot pathogens

Fifty gram leaves of Lantana, Adathoda and garlic bulbs were taken separately, washed in sterile water, disinfected with 70 per cent ethyl alcohol and then exposed to U.V. light for one hour by keeping it upside down for every 15 min. The extract was prepared by macerating using sterilized pestle and mortar with 50 ml of sterile water under aseptic condition and was filtered through clean, sterilized muslin cloth. Required quantity of the standard extract was added to molten agar medium at $45^{0}C$ and then poured into sterile Petridishes.Likewise the desired concentration of neemoil (unim) was also prepared by adding appropriate quantity directly to 100ml medium. Mycelial disc of 8 mm diameter was taken from actively growing culture of the fungus and placed on the centre of the medium Five replications were maintained for each treatment along with the control. The diameter of the fungal colony was recorded upto and when the growth in the control plates were fully covered the medium. The per cent inhibition was calculated as mentioned above.

The botanicals used for *in vitro* evaluation are presented in Table 3.

Table 3 Botanicals used for *in vitro* evaluation against leaf spot pathogens

Sl.No.	Botanicals used	Concentration (per cent)
1	Bulb extract of *Allium sativum*	5, 10, 15
2	Leaf extract of *Lantana camera*	5, 10, 15
3	Leaf extract of *Adhatoda vasica*	5, 10, 15
4	Neem oil (Unim)	0.1, 0.2, 0.3

3.11.3 *In vitro* evaluation of fungal antagonists against leaf spot pathogens

Twenty ml of medium was transferred into sterilized Petridishes. After solidification of medium, mycelial discs of 8 mm diameter was cut from actively growing culture of the fungal pathogen and placed in the centre of one half of the Petridish. On second day, the fungal antagonist was similarly transferred and placed at the centre of the other half of the same Petridish. Five replications were maintained for each and the pathogen and antagonist grown as monocultures served as control. The growth measurements were taken at regular intervals after 24h of inoculation.

The nature of antagonistic reaction against pathogen was assessed by following method of Purkayastha and Battacharya (1982).

Type of reaction	
Homogenous	: Free intermingling of hyphae
Over growth	: Pathogen over grown by antagonists
Cessation of growth	: Cessation of growth at line of contact
Aversion	: Development of clear zone of inhibition

For selecting the most efficient antagonists against pathogens, a modified Antagonistic Index (AI) suggested by Kasinathan (1998) was followed. According to this, antagonist index was calculated using the formula given below:

AI = PI x TIME x CB x IZ

where PI = Per cent inhibition

TIME = Time taken by the antagonist/pathogen to overgrow after the contact of antagonist/both

CB = Colonization behaviour of antagonist on pathogen

IZ = Inhibition zone

3.11.4 Evaluation of bacterial antagonist against leaf spot pathogens

The following methods of inoculation were adopted for testing the effect of bacterial antagonist against different leaf spot pathogens.

i) Streak method: A mycelial disc of pathogen of 8 mm size was inoculated at the centre of Petridish 48h prior to inoculation of bacterium. The bacterial isolate was inoculated as a line of streak on either size of the pathogen.

ii) Ring method: The mycelial disc of pathogen of 8mm size was inoculated at the centre of the Petridish and bacterial culture was inoculated as a ring of 2.5cm around the disc.

iii) Square method: Mycelial disc of pathogen of 8mm size was inoculated at the centre of the Petridish and bacterial culture was streaked so as to get a square shaped growth around the pathogen.

The Petridishes were incubated at room temperature and observations on growth of pathogen were taken at regular intervals. The per cent inhibition of mycelial growth of pathogen over control was calculated by the same method as given in 3.11.2.

The following antagonists were used for *in vitro* evaluation. (Table 4)

Table 4 Antagonists used for *in vitro* evaluation against leaf spot pathogens

Sl.No.	Antagonist used
1	*Trichoderma viride*
2	*Trichoderma harzianum*
3	*Aspergillus niger*
4	*Chaetomium globosum*
5	*Pseudomonas fluorescens*

3.12 Evaluation of phytotoxic effect of copper fungicides on ivy gourd

Phytotoxic effect of copper fungicides (copper oxychloride and copper hydroxide) on ivygourd leaves were tested by spraying of three concentrations (0.1, 0.2 and 0.3 per cent) of both fungicides on plants raised in polybags. The plants sprayed with water served as control.

3.13 Pot culture experiment for evaluating the effectiveness of fungicides, botanicals and antagonists against leaf spot diseases

A pot culture experiment was laid out to study the efficacy of selected fungicides, botanicals and antagonists against leaf spot diseases. The experiment was carried out during October to December 2002.

Earthern pots of size 9 inches x 9 inches were filled with potting mixture containing soil, sand and cowdung at the ratio 1:1:1. Ivy gourd cuttings were planted in pots and sprayed with appropriate concentrations of fungicides, botanicals and antagonistic organisms soon after the first appearance of the disease. Three sprays were given at 15 days interval. Disease incidence and severity were recorded before spraying and every 10^{th} and 15^{th} day after each spraying. The concentration of fungicides and botanicals found to be the most effective in *in vitro* evaluation were taken for pot culture experiment. Plants sprayed with water were maintained as control. The details of the experiment are as follows.

Variety	- Popular local variety
Treatments	- 12
Number of plants / treatment	- 4
Design	- CRD
Replication	- 3

Details of treatments used in pot culture studies are presented in the Table 5.

Table5 Details of treatments used in pot culture experiment

Treatments	Chemicals\Botanicals\Antagonists used	Concentration (per cent)
T_1	Copper hydroxide	0.2
T_2	Copper oxychloride	0.2
T_3	Captan	0.2
T_4	Mancozeb	0.2
T_5	Adathoda leaf extract	10
T_6	Neem oil	0.2
T_7	Lantana leaf extract	10
T_8	Garlic bulb extract	10
T_9	*Trichoderma viride*	0.4
T_{10}	*Trichoderma harzianum*	0.4
T_{11}	*Pseudomonas fluorescens*	0.2
T_{12}	Control	-

3.14 Field evaluation for the efficacy of selected fungicides, botanicals and antagonists against leaf spot diseases

In order to find out the effectiveness of selected fungicides, botanicals and antagonists in reducing the severity of different leaf spot diseases under natural conditions, a field experiment was laid out at College of Horticulture, Vellanikkara. Details of the field experiment were as follows:

Variety - Popular local variety
Treatments - 7 (4 plants/ treatment)
Design - RBD
Replications - 3
Plot size - 2 m x 2 m

Land was ploughed thoroughly, weeds and stubbles were removed. Two cuttings were planted in each ring at desired spacing. Crop received the respective cultural and manurial practices as recommended by Package of Practices, KAU (1996). The treatments which were found most effective against all the leaf spot organisms in *in vitro* evaluation and pot culture studies were selected for the field evaluation. The treatment details are presented in Table 6.

Table 6 Details of treatments used in field experiment

Treatments	Chemicals\Botanicals\Antagonists used	Concentration (per cent)
T_1	Copper hydroxide	0.2
T_2	Copper oxychloride	0.2
T_3	Neem oil	0.2
T_4	Garlic bulb extract	10
T_5	*Trichoderma viride*	0.4
T_6	*Pseudomonas fluorescens*	0.2
T_7	Control	-

Spraying in field was started soon after the first appearance of disease. Spraying with different fungicides, botanicals and antagonistic organisms were carried out as explained in 3.13. Per cent disease incidence and severity were also recorded.

STATISTICAL ANALYSIS

Analysis of variance was performed on the data collected in various experiments using the statistical package MSTATC (Freed, 1986). Multiple comparison among treatment means were done using DMRT.

Results

4. RESULTS

Investigations on various aspects of leaf spots diseases of such as etiology, symptomatology, varietal reaction, seasonal occurrence and management of disease were carried out and the results are presented below.

4.1 Isolation of pathogens associated with leaf spot diseases

Isolation of pathogens causing different types of leaf spot diseases on ivy gourd plants collected from three locations, *viz.*, Vellanikkara, Cherumkuzhi and Elanad showed two types of fungal colonies on Potato Dextrose Agar medium and were found belonging to *Colletotrichum* sp. and *Alternaria* sp. However, the infected leaves showing white spots yielded fungal growth only in Carrot Leaf Extract Agar and Coccinia Leaf Extract Agar media which was found to be *Cercospora* sp. All these fungi were identified up to species level based on cultural and morphological characters described later in this chapter.

Isolation of pathogens from infected fruits collected from the above three locations showed one type of fungal colony and one type of yellow bacterial colony on PDA medium. Sunken reddish brown spots showing fungal colony was found belonging to *Colletotrichum* sp. similar to that obtained from infected leaves whereas the scabby spots yielded yellow bacterial colony.

4.2 Pathogenicity

The pathogenicity of different isolates of pathogen associated with leaf spot diseases was proved by artificial inoculation under *in vitro* and *in vivo* conditions.

4.2.1 *In vitro* condition

On artificial inoculation of three different pathogens on leaves, typical spot symptoms were observed within seven to eight days of inoculation. Pathogenicity test conducted with the organism isolated from infected fruits also showed typical symptoms of fungal and bacterial infection. However, only the infection due to fungal pathogen was considered for further studies.

Reisolation of the pathogen from the infected portions showed typical original type fungal colonies on the media used.

4.2.2 *In vivo* condition

In this study also the inoculated leaves developed typical leaf spot symptoms within eight to nine days of inoculation. The pathogens reisolated from the infected portion were similar to the colonies of original culture.

Through the pathogenicity tests, it was proved that *Cercospora* sp., *Colletotrichum* sp. and *Alternaria* sp. were the pathogens causing different leaf spot diseases in ivy gourd.

4.3 Symptomatology

Studies on symptomatology of the diseases caused by the three different pathogens viz., *Cercospora* sp., *Colletotrichum* sp. and *Alternaria* sp. on the foliage of ivy gourd under natural and artificial conditions showed that, type of symptoms varied with different pathogens; however, no variation in symptoms were observed between three different locations. Similarly, the study on symptomatology of fruit infection also showed no difference in symptoms due to difference in locations.

4.3.1 Symptomatology of *Cercospora* leaf spot under natural conditions

Incidence of *Cercospora* leaf spot was severe on middle aged leaves. At first, water soaked spots appeared on lower surface of leaf and the corresponding upper surface showed chlorotic discolouration. Later, it turned to white necrotic circular spots with prominent yellow halo on upper surface and water soaked area around the spot on lower surface of the leaves. The spots remained as distinct white necrotic area and did not coalesce. In advanced stage, the spots became dry and papery and showed black dots at the centre which were the conidiophores of the organism. During severe infection, pathogen produced 20-25 spots of 1-2 mm diameter on each infected leaf (Plate IA).

4.3.2 Symptomatology of *Colletotrichum* leaf spot under natural conditions

The leaf spots caused by *Colletotrichum* sp. initiated as water soaked areas on lower surface and chlorotic spots on the corresponding upper surface of the leaves. It developed as circular spot with yellow halo which was very clear when held against the light. Later it turned to brown necrotic spots with brown margin on both sides of leaf surrounded by yellow halo. In advanced stage, spots coalesced to form large brown necrotic lesions. Occasionally shot hole symptom was noticed. Middle aged leaves were attacked severely and spots were of 1-2.5 mm in diameter. The fungus produced its fruiting body, acervuli at the centre of the spots which appeared as black dots. During severe infection, 25-30 spots were seen on single leaf (Plate IB).

4.3.3 Symptomatology of *Alternaria* leaf spot under natural conditions

Symptoms of *Alternaria* leaf spot started from the margins of the leaves as water soaked lesions with clear yellow halo on both surfaces. Later turned to brown with characteristic concentric zonations and finally the infected area changed to dark brown, dry and papery. Usually the lesions coalesced affecting larger leaf area. Older leaves were found more susceptible to the infection. The diameter of spot ranged from 3 to 5 mm (Plate IC).

4.3.4 Symptomatology of combined infection of *Cercospora* sp. and *Colletotrichum* sp. under natural conditions

In case of combined infection of *Cercospora* sp. and *Colletotrichum* sp., spots appeared as circular reddish brown with yellow halo on upper surface and water soaked lesion on lower surface. The size of the spots were found to be 1-4 mm and black dots were present at the centre which were either conidiophores of *Cercospora* sp. or acervuli of *Colletotrichum* sp. During serious infection 25-30 spots were produced on each leaf (Plate ID).

4.3.5 Symptoms on fruits

The fruits were infected by *Colletotrichum* sp. and produced circular, sunken, reddish brown spots surrounded by water soaked area, having diameter of 1.5-2.5 mm.(Plate IE).

4.4 Symptomatology on artificial inoculation

Under artificial inoculation, it was found that the symptoms produced by the respective pathogens were almost similar to those produced in the natural conditions. However, slight variation was observed in case of *Colletotrichum* sp. which produced a clear brown margin around the spots. In plants inoculated with *Cercospora* sp. the initial white spots later turned to dark brown, with yellow halo.

4.5 Histopathological studies

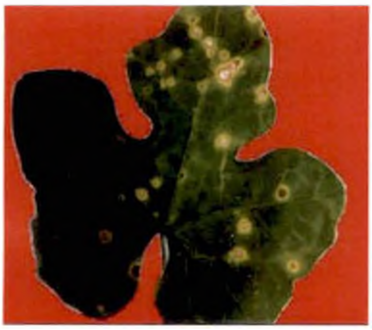
Thin sections of different leaf spot-affected areas were taken to study the internal symptoms caused by the pathogen in the host tissues. In the case of *Cercospora* infection, severe distortion of tissues were observed up to mesophyll layer. Conidiophores were found to initiate from the palisade tissues (Plate IF) whereas *Colletotrichum* infection caused only slight damage of the epidermal cells. With respect to *Alternaria* infection, damage was observed on epidermis in which a few sub epidermal cells were found to be destroyed. Combined infection of *Cercospora* sp. and *Colletotrichum* sp. affected the distribution pattern of

A) *Cercospora* leaf spot

a) Upper surface

b) Lower surface

B) *Colletotrichum* leaf spot



C) *Alternaria* leaf spot

D) Combined infection of *C. cocciniae* and *C. gloeosporioides*

E) Symptom on fruits

F) Leaf section showing conidiophores

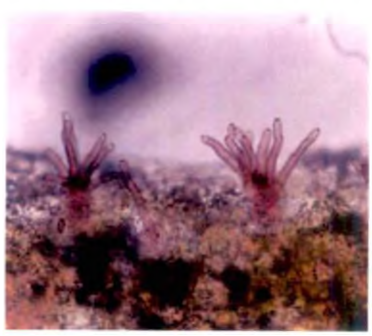


Plate 1. Symptomatology

chloroplasts and also caused disintegration of chlorophyll grains. During the advanced stage, necrosis and death of complete cells were noticed in all types of infections.

4.6 Identification of pathogens

The pathogens causing different leaf spots were identified based on cultural and morphological characters.

4.6.1 Cultural and morphological characters of the pathogen

Cultural and morphological characters of the three pathogens on selective medium were studied in detail and based on that these organisms were identified. These characters of different pathogens from three locations were almost similar with negligible variations. Details of morphological characters are given in Table 7.

4.6.1.1 Cercospora sp.

Carrot Leaf Extract Agar and Coccinia Leaf Extract Agar media were used for studying the characters of *Cercospora sp.*. The cultural characters observed were almost same in both media. Colony was slow growing and completed 9 cm diameter growth in eight days at room temperature. Initially a sparse white mycelial growth was seen which turned to pink colour on the third day of incubation. Later the basal portion of the fungal colony turned brownish black with slight pink colour on the upper surface. Black fruiting structures were present on the growth, and reverse of the colony appeared greenish black. Conidial formation was not observed in media. Fusion between undifferentiated vegetative hyphae, i.e., anastomosis was observed in the culture of *Cercospora* sp.

Hyphae branched, brown with 3.8-5.8 µm width and septate at intervals of 11.6-34.8 µm (Plate 2A).Measurements of conidia and conidiophores were taken from host tissues. Conidiophores abundant on lower side of the leaf, fasiculate, dark brown unbranched, flexous, smooth, geniculate, scars distinct and

46.7-155.6 μm x 3.9 μm with 3-6 septa. Conidia solitary, hyaline, cylindrical, apex slightly tapered, 23.3-93.4μm x 3.9 μm with 4-12 septa (Fig. 1).

Based on the above characters, the organism was identified as *Cercospora cocciniae* Munjal, Lall and Chona.

4.6.1.2 Colletotrichum sp.

Potato Dextrose Agar medium was used for studying the characters of *Colletotrichum* sp. Colony was fast growing, attained 9 cm diameter growth in 6 days at the room temperature. Pinkish white growth on the medium was observed two days after incubation, mycelial growth turned brown with greyish white colour on the upper surface. Finally the growth changed to dark brown with pink pigmentation. Reverse of the colony black, acervuli were present after 10 days of incubation (Plate 2 B).

Hyphae branched, hyaline with 3.8 μm width and septate at an interval of 11.6-19.4 μm. Conidia hyaline, cylindrical with both ends round, aseptate, oil globules present, 11.7-15.6 μm x 3.9 μm. Setae dark brown with 1-2 septa, swollen at the base and tapering at the apex (Fig. 2).

Based on the characters, the organism was identified as *Colletotrichum gloeosporiodes* (Penz.) Sacc. The pathogen obtained from fruits also showed the same characters and identified as *C. gloeosporiodes* (Penz.) Sacc.

4.6.1.3 Alternaria sp.

Cultural and morphological characters of *Alternaria* sp. were studied on PDA medium. It showed fast growth on PDA and completed 9 cm diameter growth in seven days at the room temperature. The pure culture of the fungus first showed whitish growth, later lower portion of the colony turned dark brownish black colour. The mycelial growth had a velvety appearance with dark purplish tinge on the upper side and the reverse of the colony almost black.

Table 7 Morphological characters of leaf spot pathogens

Organism	Hypha		*Conidia*						Conidiophores		
	Width (µm)	Distance between two septa	Length (µm)	Width (µm)	Number of septum		Length of beak (µm)	Number of septum (beak)	Length (µm)	Width (µm)	Number of septum
					Transverse	Longitudinal					
1.a) *Cercospora* sp. (VKA)	3.8-5.8	11.6-34.8	23.3-93.4	3.9	4-12				46.7-155.6	3.9	3-6
b) *Cercospora* sp. (CHKI)	3.8-5.8	11.6-34.8	23.3-97.3	3.9	6-14				46.7-155.6	3.9	3-6
c) *Cercospora* sp. (ELD)	3.8-5.8	11.6-34.8	23.3-89.5	3.9	6-12				42.8-151.7	3.9	3-6
2 a) *C. gloeosporioides* (VKA)	3.8	11.6-19.4	11.7-15.6	3.9							
b)*C.gloeosporioides* (CHKI)	3.8	11.6-19.4	11.7-15.6	3.9							
c) *C. gloeosporioides* (ELD)	3.8	11.6-19.4	7.8-15.6	3.9							
3.a) *A. alternata* (VKA)	3.8-5.8	15.4-30.9	19.5-31.2	7.8-15.6	3-5	1-3	3.9-27.3	1-2			
b) *A. alternata* (CHKI)	3.8-5.8	15.4-30.9	23.4-35.1	7.8-15.6	3-6	1-3	3.9-27.3	1-2			
c) *A. alternata* (ELD)	3.8-5.8	15.4-30.9	19.5-31.2	7.8-15.6	3-5	1-3	3.9-23.4	1-2			

VKA - Vellanikkara
CHKI - Cherumkuzhi
ELD - Elanad
Average of 50 measurements

A) *C. cocciniae*

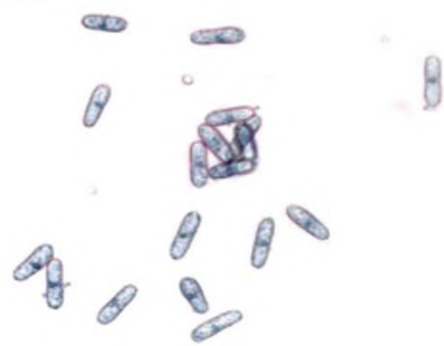
a) Conidiophore (400x)

b) Conidia (1000x)

c) Hyphae showing anastomosis (400x)

B) *C. gloeosporioides*

a) Acervulus (400x)



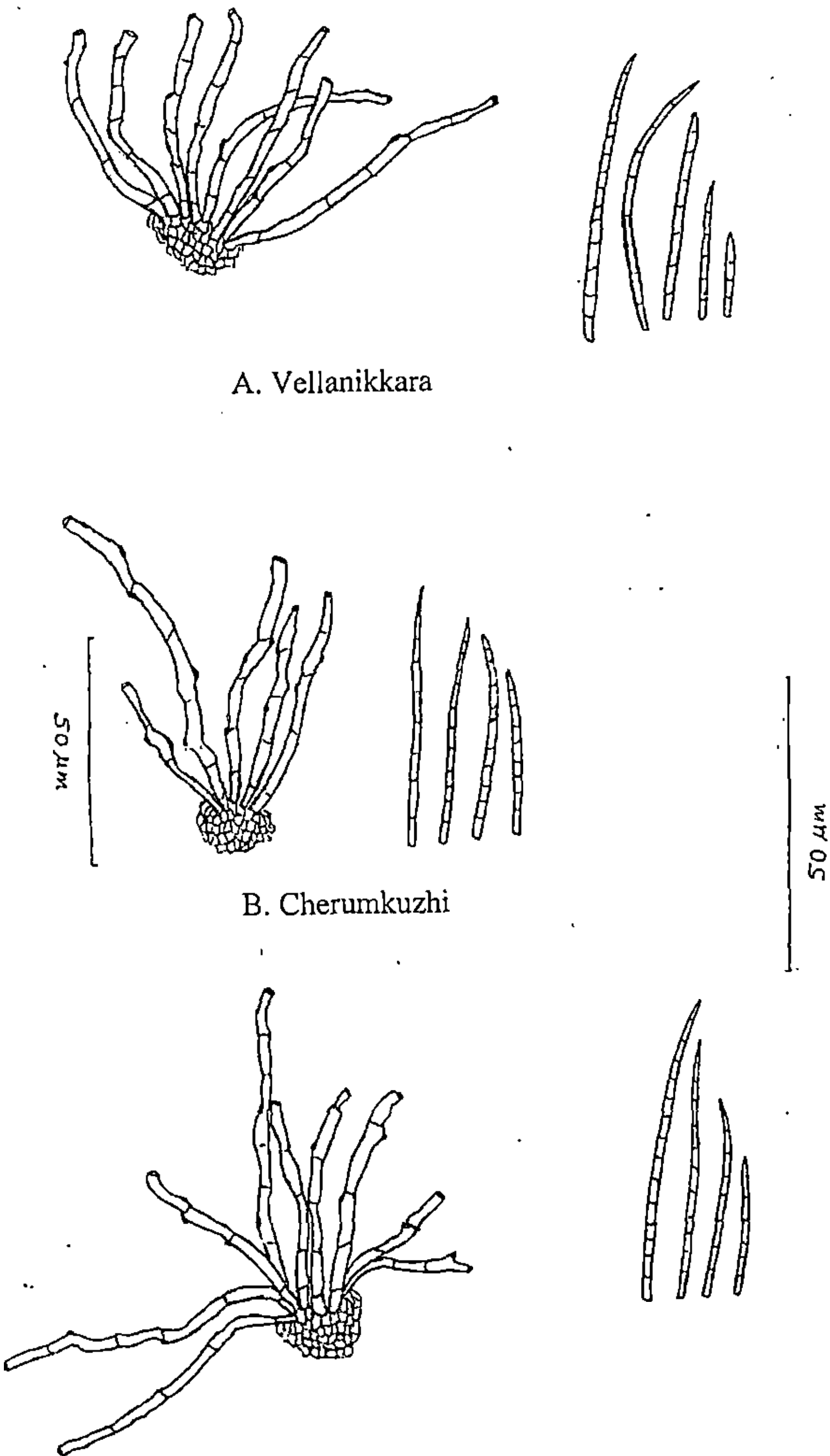
b) Conidia (400x)

C) *A. alternata*

a) Conidia (1000x)

Plate 2. Cultural and morphological characters of pathogens

Fig 1. Conidiophores and conidia of *Cercospora cocciniae* from different locations



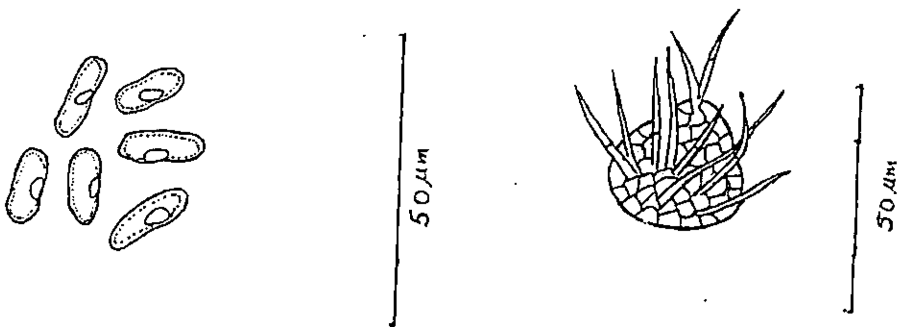
A. Vellanikkara

B. Cherumkuzhi

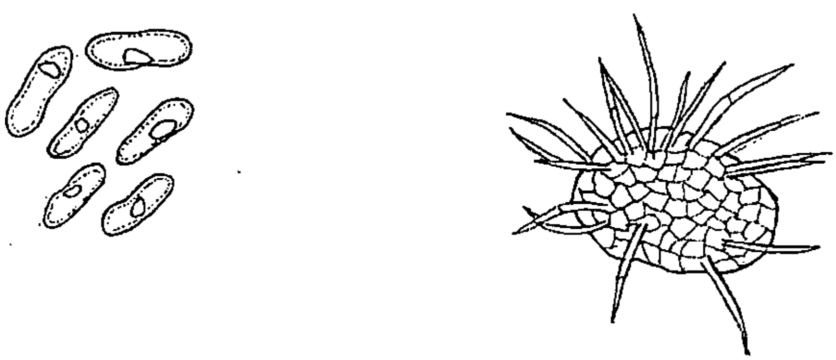
C. Elanad

Fig 2 Conidia and Acervulus of *Colletotrichum gloeosporioides*

A. Vellanikkara



B. Cherumkuzhi



C. Elanad

Hyphae branched, initially hyaline, turned brownish black with 3.8-5.8 µm width, septate at 15.4-30.9 µm intervals, condiophores were seen singly or in groups. Conidia formed in chain or sometimes free, straight, obclavate, smooth, rostrate, the basal cell rounded, pale to dark brown, 19.5-31.2 µm x 7.8-15.6 µm and with 3-5 transverse and 1-3 longitudinal septa(Fig.3). Beak prominent, measuring 3.9-27.3 µm length with 1-2 transverse septa (Plate 2 C).

Based on these characters, the organism was identified as *Alternata alternata* Fr. Keissler.

4.7 Studies on conidial germination of leaf spot pathogens

Spore suspension of *C. cocciniae, C. gloeosporioides* and *A. alternata* were prepared separately, incubated and observed for the germination at one hour interval. The details are presented in Table 8.

Spore suspension of *C. cocciniae* was prepared from naturally infected leaves. Germination of spores started at two hours after incubation (HAI) and recorded 12 per cent germination. A rapid increase in germination was observed in the subsequent intervals and attained cent per cent germination at seven hours after incubation. Germ tube formed from all cells of conidia, but more frequently from the terminal cells and developed into hypha (Plate 3 A).

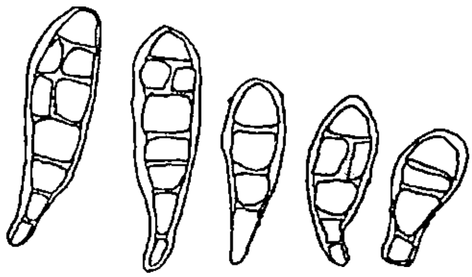
Spores of *C. gloeosporioides* showed 8 per cent germination at three hours after incubation and recorded cent per cent germination at nine hours after incubation. Conidia germinated from both ends and produced germ tube with appressorium at the end. The appressorium appeared irregular in shape and brownish black in colour (Plate 3 B).

Spores of *A. alternata* germinated at three hours after incubation and recorded only 6.0 per cent germination. It recorded cent per cent germination at ten hours after incubation. All cells of the conidium germinated by producing germ tube which elongated to form hypha (Plate 3 C).

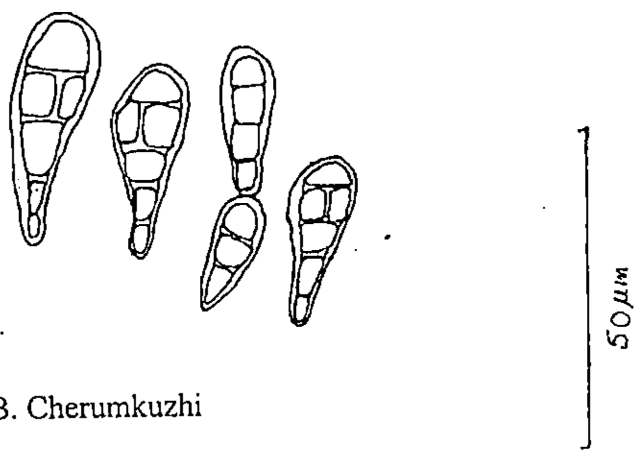
Table 8. Per cent germination of spores of different pathogens

Sl. No	Organisms	Hours after incubation (HAI)									
		1	2	3	4	5	6	7	8	9	10
1	*C. cocciniae*	-	12	20	33.3	66.6	93.3	100			
2	*C. gloeosporioides*	-	-	8	24	40	60.5	72	89.5	100	
3	*A. alternata*	-	-	6	20	33.3	70	78.5	86.6	92	100

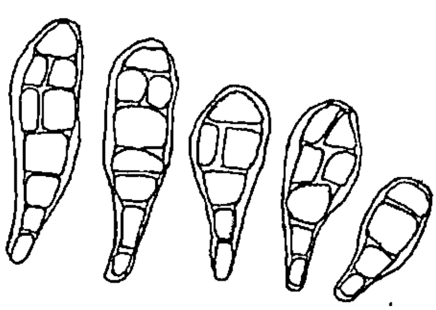
Fig 3. Conidia of *Alternaria alternata* from different locations



A. Vellanikkara

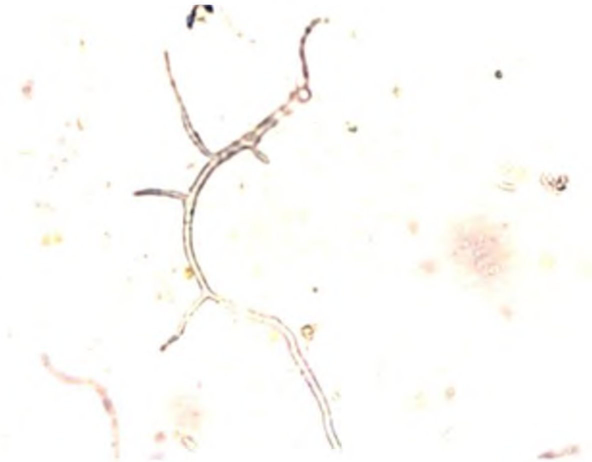


B. Cherumkuzhi

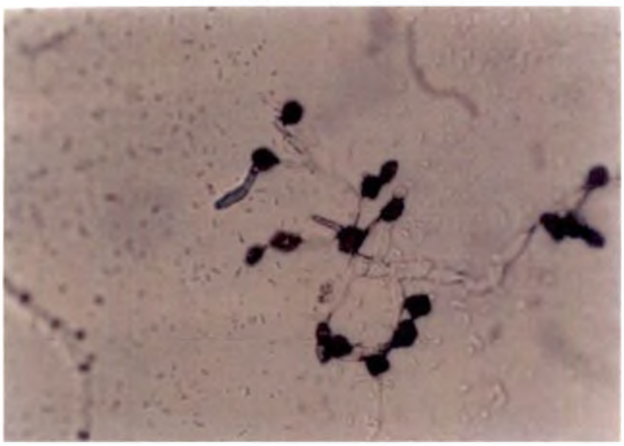


C. Elanad

A) *C. cocciniae*



B) *C. gloeosporioides*



Germinating conidia with appressorium

C) *A. alternata*

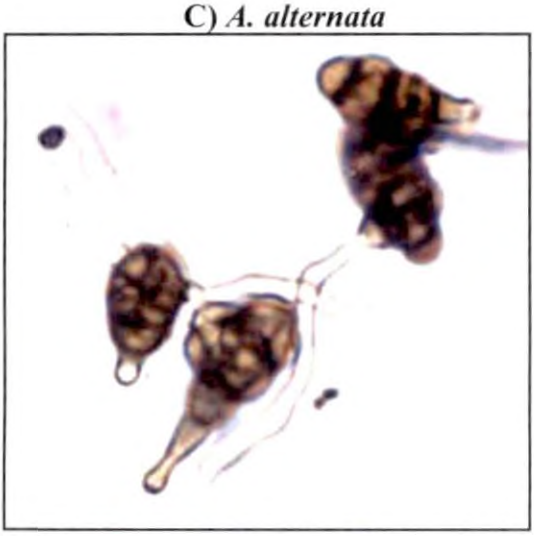


Plate 3. Conidial germination of different pathogens

4.8 Effect of toxin on disease development

In order to find out the effect of toxins of *C. cocciniae*, *C. gloeosporioides* and *A. alternata* on disease development, the leaves of ivy gourd plants were spotted separately with the endotoxin and exotoxin of these pathogens. The effect of toxin was first noticed on plants spotted separately with the exotoxin and endotoxin of *C. cocciniae* and inoculated leaves showed yellowing followed by watersoaked brown lesions of five days after incubation. Gradually, the symptoms spread to older and younger leaves and complete yellowing of the plants occurred. In case of *C. gloeosporioides*, the leaves spotted with endotoxin showed yellowing and water soaked lesions six days after inoculation, whereas, in *A. alternata*, a slight yellow discolouration was observed on those spotted with endotoxin after seven days of inoculation. No symptoms were observed in plants treated with exotoxins of *C. gloeosporioides* and *A. alternata*.

4.9 Evaluation of ivy gourd genotypes for resistance to leaf spot diseases

Nineteen ivy gourd genotypes were screened against leaf spot diseases under natural condition. The results of the experiment are furnished in Table 9. From the data, it was found that the genotypes showed variation in resistance to leaf spot diseases. Among the nineteen genotypes, none of them were found completely free of disease. However, eight genotypes, CG 8, CG 9, CG 10, CG 11, CG 19, CG 23, CG 27 and CG 31 showed CI value ranged from 0.4 to 3.6 and were found to be highly resistant to leaf spot diseases. Among these, CG 23 showed lowest CI value of 0.4. Like wise, in case of CG 3, CG 5, CG 6 and CG 20, the CI values ranged from 4.8 to 8 and were categorized as resistant ones. Only two genotypes showed moderately resistant reactions and rest of them were found to be moderately susceptible and CG 32, the local variety was found susceptible to leaf spot disease, recording the maximum CI value of 42.6.

4.10 Seasonal influence on the incidence of leaf spot diseases

The influence of season on the incidence of different leaf spots on ivy gourd was observed by recording per cent disease incidence and severity from the

Table 9 Evaluation of ivy gourd genotype for resistance against leaf spot diseases

Sl. No.	Genotypes	Per cent disease incidence	Per cent disease severity	Coefficient of Infection	Disease reaction
1	CG I	50	20.6	10.3	MR
2	CG 3	40	15.5	6.2	R
3	CG 5	40	20	8.0	R
4	CG 6	25	19	4.8	R
5	CG 7	100	28.6	28.6	MS
6	CG 8	20	18	3.6	HR
7	CG 9	9.1	6.5	0.6	HR
8	CG 10	9.3	9.5	0.8	HR
9	CG 11	16.7	20	3.3	HR
10	CG 17	100	28	28	MS
11	CG 18	100	26	26	MS
12	CG 19	16.7	12	2.0	HR
13	CG 20	25	21	5.3	R
14	CG 21	50	24	12	MR
15	CG 23	8.3	5	0.4	HR
16	CG 26	66.7	31	20.7	MS
17	CG 27	9.4	6.5	0.6	HR
18	CG 31	16.7	16	2.7	HR
19	CG 32 (Local variety)	100	42.6	42.6	S

HR - Highly resistant

R - Resistant

MR - Moderately resistant

MS - Moderately susceptible

S - Susceptible

three locations surveyed. During the survey it was noticed that the leaf spots appeared individually and also in combination especially in case of *C. cocciniae* and *C. gloeosporioides*. The data are presented in the Table 10.

From the data it was observed that leaf spot caused by *C. cocciniae* was the most serious disease observed in all the three locations. Severe infection of *Cercospora* leaf spot alone was observed during June-July months, of which the maximum incidence (100 per cent) and severity (55.20 per cent) was observed in June. It was also noticed that the maximum infection was observed in Cherumkuzhi areas when compared to Vellanikkara and Elanad. Even though the individual Cercospora leaf spot infection was not observed during August, combined infection of *C. cocciniae* and *C. gloeosporioides* were found severe in all the three locations during this period, recording cent per cent incidence and 49.74 to 59.40 per cent severity. Leaf spot infection was found to decrease in the subsequent months. During this period, minimum temperature of 23.10-23.3°C and maximum temperature varied from 28.9°-30.0°C with a mean temperature ranged from 26.5°-26.7°C. It was also observed that relative humidity and rainfall were also high ranging 84-86 per cent and 354.2-533.5 mm respectively (Table 11).

Individual infection of *C. gloeosporioides* was observed only during the month of June and was observed only at Vellanikkara and Elanad locations. However, the combined infection with *C. cocciniae* was observed in the subsequent months in all the locations.

From the data presented in Table 10, it was observed that *Alternaria* infection was comparatively low in Elanad areas as compared to other two locations and the severe infection was observed in Cherumkuzhi areas. Alternaria leaf spot was more prevalent during the summer months (February to March) showing 38-69.04 per cent incidence and 23.66-33.20 per cent disease severity in Vellanikkara and Cherumkuzhi areas. During these months, minimum temperature of 23.6°-24.1°C and the maximum temperature of 33.2-34.7°C were recorded with a mean temperature of 29.2°-29.4°C. The relative humidity ranged from 63-67 per cent and the total rainfall during this period was 94.8-162.1 mm. However, a severe infection of this pathogen was noticed in Cherumkuzhi area during

Table10 Per cent disease incidence and severity of various leaf spots of ivy gourd in Vellanikkara, Cherumkuzhi and Elanad during June 2002 to May 2003

	Cercospora leaf spot						Colletotricum leaf spot						Combined infection						Alternaria leaf spot					
Period	VKA		CHKI		ELD		VKA		CHKI		ELD		VKA		CHKI		ELD		VKA		CHKI		ELD	
	PDI	PDS	PDI	PDS	PDI	PDS	PDI	PDS	PDI	PDS	PDI	PDS	PDI	PDS	PDI	PDS	PDI	PDS	PDI	PDS	PDI	PDS	PDI	PDS
June 02	100	34.20	100	55.20	100	39	19.56	12.66	0	0	75.00	34.00	0	0	0	0	0	0	0	0	0	0	0	0
July	0	0	100	53.80	0	0	0	0	0	0	0	0	100	56.30	0	0	100	45.20	0	0	0	0	0	0
Aug	0	0	0	0	0	0	0	0	0	0	0	0	100	49.74	100	50.30	100	59.40	0	0	0	0	0	0
Sept	0	0	0	0	0	0	0	0	0	0	0	0	100	20.60	40.00	47.00	100	57.40	0	0	100	47.20	0	0
Oct	0	0	0	0	100	47.20	0	0	0	0	0	0	100	24.10	100	45.80	0	0	0	0	100	31.60	0	0
Nov	0	0	0	0	100	52.20	0	0	0	0	0	0	100	28.14	80.00	34.40	0	0	0	0	0	0	0	0
Dec	0	0	0	0	100	42.20	0	0	0	0	0	0	100	35.86	75.00	28.40	0	0	0	0	0	0	0	0
Jan 03	66.66	20.33	0	0	100	40.40	0	0	0	0	0	0	0	0	60.00	25.60	0	0	12.73	0.87	0	0	0	0
Feb	73.81	24.05	0	0	100	30.40	0	0	0	0	0	0	0	0	0	0	0	0	50	23.66	38	28.20	23.76	8.40
Mar	85.71	24.90	0	0	100	29.20	0	0	0	0	0	0	0	0	0	0	0	0	69.04	23.66	44	33.20	32.33	14.80
Apr	92.86	28.52	28.00	24.80	100	36.60	0	0	0	0	0	0	0	0	0	0	0	0	42.86	17.48	0	0	0	0
May	60.00	21.00	92.00	42.40	100	38.00	12.40	8.22	50	18.60	60.00	12.12	0.00	0	0	0	0	0	21.84	4.26	0	0	0	0

VKA - Vellanikkara
CHKI - Cherumkuzhi
ELD - Elanad
PDI-Per cent disease incidence
PDS-Per cent disease severity

Table 11 Weather data at monthly intervals from June2002 to May 2003

Period	Temperature (°C)			Mean Relative Humidity (Per cent)	Average Rainfall (mm)
	Minimum	Maximum	Mean		
June2002	23.3	30.0	26.7	86	533.5
July	23.1	29.8	26.5	84	354.2
August	22.9	28.9	25.9	86	506.6
September	23.0	31.1	27.0	77	125.0
October	23.2	30.8	27.0	83	387.7
November	23.4	31.8	27.6	71	22.1
December	22.1	32.3	27.2	45	0.0
January2003	22.9	33.2	28.0	50	0.0
February	23.6	34.7	29.2	63	162.1
March	24.1	34.6	29.4	67	94.8
April	25.0	34.6	29.8	72	23.8
May	25.0	34.0	29.5	72	40.3

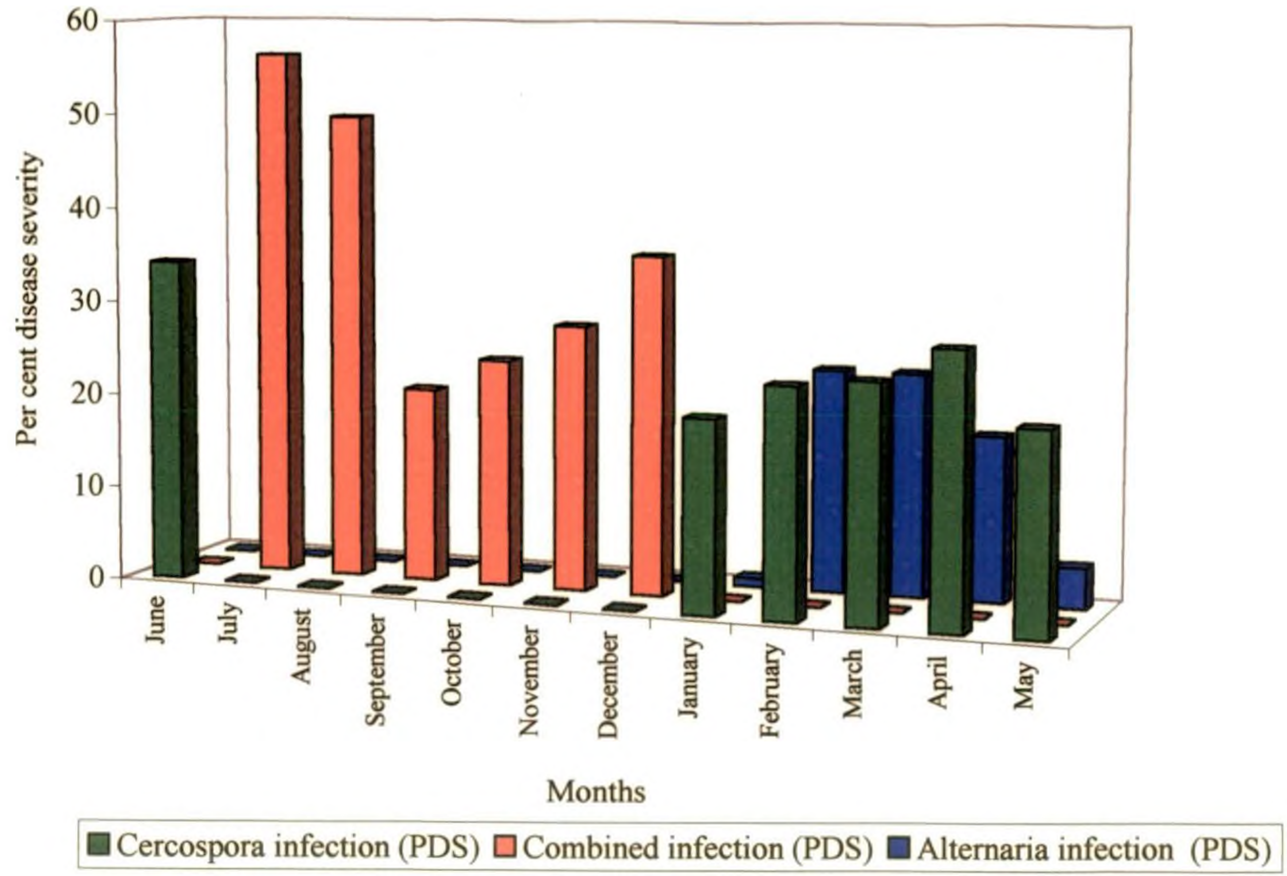


Fig. 4. Seasonal influence on severity of leaf spot diseases of Ivy gourd at Vellanikkara

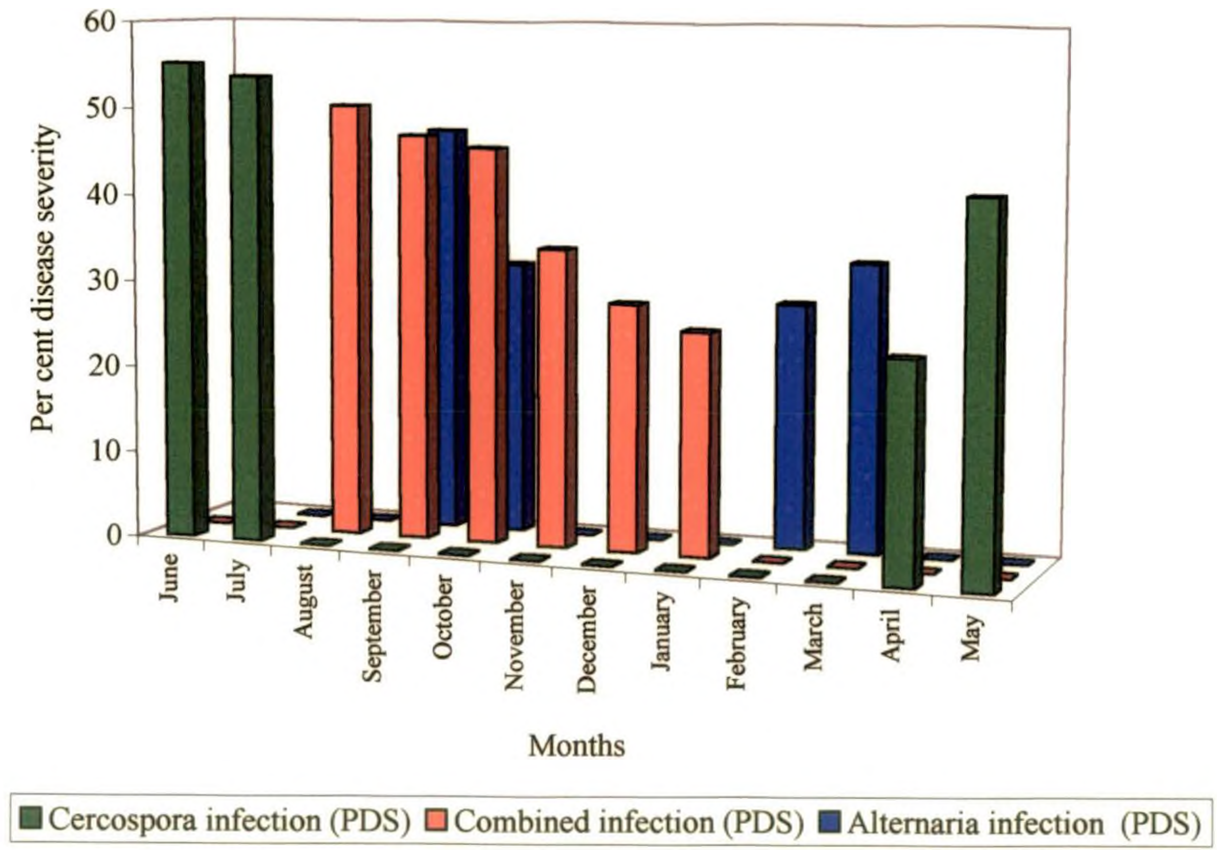


Fig. 5. Seasonal influence on severity of leaf spot diseases of Ivy gourd at Cherumkuzhi

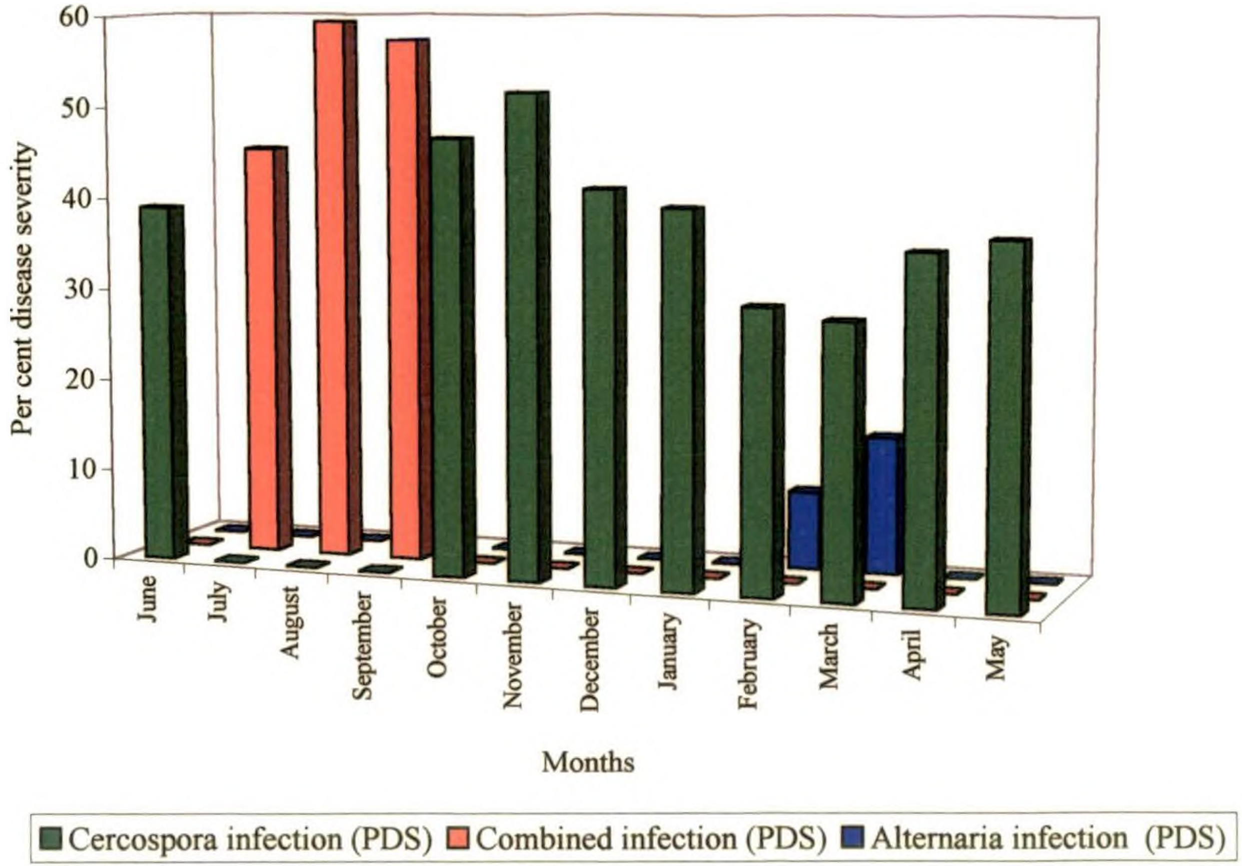


Fig. 6. Seasonal influence on severity of leaf spot diseases of Ivy gourd at Elanad

September-October months during which relative humidity and rainfall was comparatively less as compared to previous months.(Fig 4,5&6)

4.11 Management of leaf spot diseases

The efficacy of chemicals, botanicals and antagonistic organisms against the three leaf spot pathogens were evaluated under *in vitro* condition. The results of the experiments are given in Table 12-14.

4.11.1 *In vitro* evaluation of fungicides and botanicals against *C. cocciniae*

4.11.1.1 Fungicides

The efficacy of different fungicides at various concentration on the growth of *C. cocciniae* is given in Table 12. From the data, it is evident that the treatments differed significantly with each other in their efficiency in inhibiting the growth of *C. cocciniae*. All fungicides recorded maximum inhibition at 0.2 per cent concentration.

Among the fungicides, captan, 0.2 per cent concentration was found to be most inhibitory to the pathogen, recording 13.2 mm growth against full growth in the control at eight days after incubation, showing 85.32 per cent inhibition over control. It was on par with captan 0.3 per cent and copper oxychloride 0.2 per cent recording 83.22 and 82.66 per cent inhibition respectively. Mancozeb, 0.1 per cent concentration showed least inhibition (55.32 per cent) to the pathogen.(Plate 4A)

4.10.1.2 Botanicals

From the results furnished in the Table 12, it was revealed that all treatments differ significantly with each other in their inhibitory effect on the growth of *C. cocciniae*. All botanicals recorded highest inhibition at 10 per cent concentration except garlic. The bulb extract of garlic at all the three concentrations showed inhibitory effect on the growth of the pathogen from the second day of incubation and recorded only 8 mm diameter till eighth day after incubation, when the control plates showed full growth of the fungus. This resulted

Table 12. *In vitro* evaluation of fungicides and botanicals against *C. cocciniae*

a. Fungicides

Sl. No.	Treatments	Concentration (Per cent)	DAI (diameter in mm)				Percent inhibition over control
			2nd	4th	6th	8th	
1	Copper hydroxide	0.1	13.1^{f}	17.4^{g}	24.9^{j}	29.4^{i}	67.33
		0.2	9.3^{hi}	10.9^{jk}	13.4^{op}	15.6^{no}	82.66
		0.3	9.3^{hi}	11.1^{jk}	14.7^{no}	17.1^{mn}	80.99
2	Copper oxychloride	0.1	12.1^{g}	18.8^{f}	24.0^{jk}	26.9^{j}	70.11
		0.2	9.3^{hi}	11.7^{jk}	17.3^{m}	20.7^{l}	76.99
		0.3	12.4^{fg}	15.3^{h}	19.9^{l}	23.5^{k}	73.88
3	Captan	0.1	9.8^{hi}	11.9^{j}	15.5^{n}	18.6^{m}	79.32
		0.2	9.0^{i}	10.0^{l}	11.9^{p}	13.2^{p}	85.32
		0.3	9.4^{hi}	10.7^{kl}	12.2^{p}	15.1^{o}	83.22
4	Mancozeb	0.1	14.4^{e}	20.9^{e}	31.0^{h}	40.2^{h}	55.32
		0.2	11.7^{g}	13.6^{v}	19.2^{l}	22.7^{k}	74.77
		0.3	10.2^{h}	15.1^{h}	30.1^{I}	30.1^{i}	66.55
Control			22.8^{a}	44.8^{a}	68.2^{a}	90.0^{a}	
b. Botanicals							
1	Lantana	5	15.4^{cd}	24.3^{cd}	43.9^{d}	57.9^{d}	35.66
		10	14.3^{e}	21.7^{e}	41.2^{ef}	52.3^{f}	43.33
		15	16.4^{b}	24.9^{cd}	48.8^{c}	61.1^{c}	32.09
2	Adathoda	5	16.0^{bc}	24.4^{cd}	40.1^{f}	60.1^{c}	33.19
		10	14.3^{e}	24.4^{cd}	37.5	51.84^{f}	42.33
		15	16.6^{b}	29.9^{b}	58.5^{b}	69.4^{b}	22.88
3	Garlic	5	8.0^{j}	8.0^{m}	8.0^{q}	8.0^{q}	91.11
		10	8.0^{j}	8.0^{m}	8.0^{q}	8.0^{q}	91.11
		15	8.0^{j}	8.0^{m}	8.0^{q}	8.0^{q}	91.11
4	Neem oil	0.1	13.1^{f}	23.9^{d}	36.7^{g}	52.7^{ef}	41.44
		0.2	12.1^{g}	19.3^{f}	29.2^{i}	45.2^{g}	49.77
		0.3	14.9^{de}	25.2^{c}	42.3^{e}	54.4^{e}	39.53
Control			22.9^{a}	44.4^{a}	69.3^{a}	90.0^{a}	

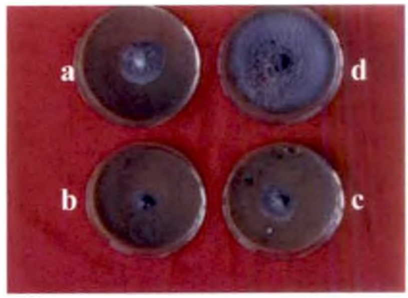
DAI - Days after inoculation

* Mean of five replications.

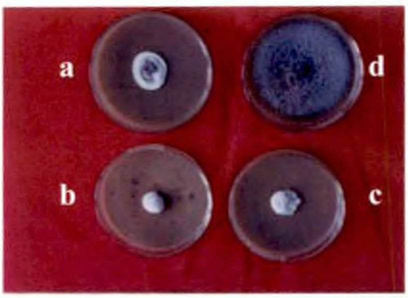
In each column figures followed by same letter do not differ significantly according to DMRT

A) Fungicides

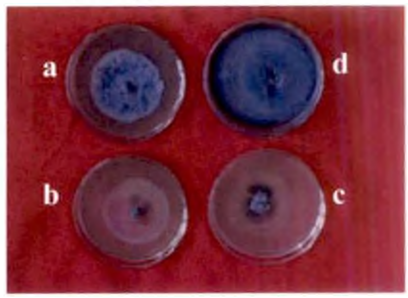
1) Copper hydroxide



2) Copper oxychloride



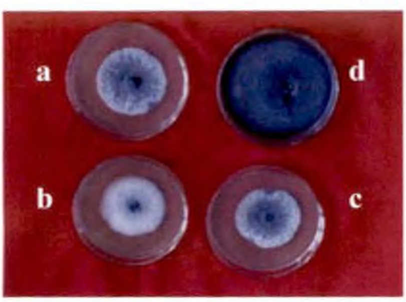
3) Captan



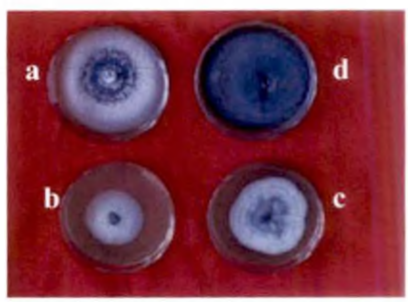
4) Mancozeb

a - 0.1 per cent concentration
b - 0.2 per cent concentration
c - 0.3 per cent concentration
d - Control

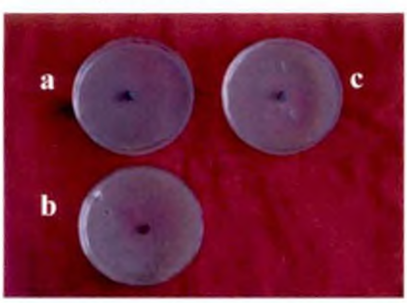
B) Botanicals



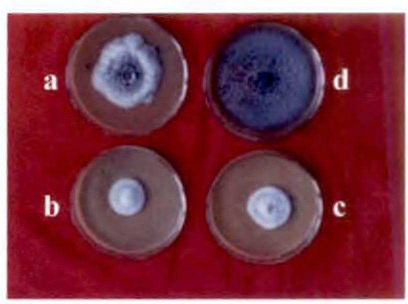
1) Lantana



2) Adathoda



3) Garlic



4) Neem oil (0.1, 0.2, 0.3 per cent)

a - 5.0 per cent concentration
b - 10.0 per cent concentration
c - 15.0 per cent concentration
d - Control

Plate 4. *In vitro* evaluation of fungicides and botanicals against *C. cocciniae*

in maximum inhibition of growth and recorded 91.11 per cent inhibition over control (Plate 4 B)

4.11.2 *In vitro* evaluation of fungicides and botanicals against *C. gloeosporioides*

4.11.2.1 Fungicides

From the data given in Table 13, it was found that the treatments differ significantly with each other in their efficiency against the growth of *C. gloeosporioides*.

The maximum growth of 90 mm in the control was recorded at six days after incubation. Copper hydroxide even at 0.2 and 0.3 per cent concentration were inhibitory to the pathogen and recorded 90 per cent inhibition over control. These concentrations were on par with 0.1 per cent copper hydroxide and also with copper oxychloride at all the three concentration which recorded a range of 89.44-89.77 per cent inhibition on the growth of the pathogen. Captan at all concentrations showed less inhibition on the growth of the fungus and recorded inhibition between 66.11 to 73.44 per cent over control. Mancozeb 0.1 per cent concentration recorded least inhibition(14.77 per cent)on the growth of pathogen, among the different concentrations of various fungicides tested.

4.11.2.2 Botanicals

The data given in Table 13 showed statistical difference among the treatments in their effectiveness on inhibiting the growth of *C. gloeosporioides*.

Among the botanicals, the 10 per cent bulb extract of garlic showed the maximum inhibition on the growth (85.66per cent) of the pathogen. It was superior than all other botanicals and was followed by 80.22 per cent inhibition over control recorded by garlic at 15 per cent concentration. Neem oil at all the three concentrations were found on par with each other and recorded growth inhibition ranging from 29.55 to 31.66 per cent over control. Adathoda at 15 per cent concentration showed the least inhibition on the growth of fungus and recorded only 15.55 per cent inhibition over control. (Plate 5B)

Table 13 *In vitro* evaluation of fungicides and botanicals against *C.gloeosporioides*

a. Fungicides

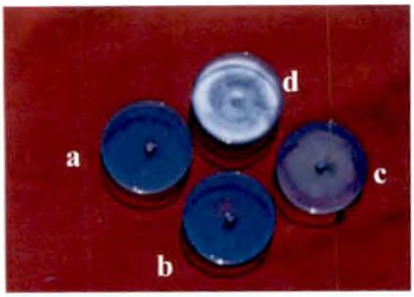
Sl. No .	Treatments	Concentration (Per cent)	DAI (diameter in mm)			Percent inhibition over control
			2nd	4th	6th	
1	Copper hydroxide	0.1	9.5^{lm}	9.5^{kl}	9.5^{n}	89.44
		0.2	9.0^{m}	9.0^{l}	9.0^{n}	90.00
		0.3	9.0^{m}	9.0^{l}	9.0^{n}	90.00
2	Copper oxychloride	0.1	9.2^{m}	9.2^{l}	9.2^{n}	89.77
		0.2	9.2^{m}	9.2^{l}	9.2^{n}	89.77
		0.3	9.5^{lm}	9.5^{kl}	9.2^{n}	89.77
3	Captan	0.1	16.3^{j}	21.8^{g}	30.5^{i}	66.11
		0.2	13.6^{k}	18.0^{i}	23.9^{k}	73.44
		0.3	15.9^{j}	19.6^{hi}	25.0^{k}	72.22
4	Mancozeb	0.1	26.0^{g}	50.9^{c}	76.7^{b}	14.77
		0.2	15.2^{j}	40.3^{f}	70.0^{cd}	22.30
		0.3	19.1^{i}	48.3^{d}	70.5^{c}	21.84
Control			36.8^{a}	65.6^{a}	90.0^{a}	
b. Botanicals						
1	Lantana	5	30.1^{e}	48.3^{d}	68.4^{de}	24.08
		10	34.3^{c}	51.44^{bc}	66.48^{f}	26.22
		15	24.8^{h}	44.8^{e}	67.1^{ef}	25.44
2	Adathoda	5	28.8^{f}	48.4^{d}	67.1^{ef}	19.99
		10	34.5^{bc}	51.7^{bc}	71.6^{c}	20.44
		15	31.6^{d}	49.7^{cd}	76.0^{b}	15.55
3	Garlic	5	12.5^{k}	21.0^{gh}	28.1^{j}	68.77
		10	10.7^{l}	11.3^{k}	12.9^{m}	85.66
		15	10.6^{l}	13.6^{j}	17.8^{l}	80.22
4	Neem oil	0.1	34.4^{c}	53.2^{b}	62.5^{gh}	30.55
		0.2	32.5^{d}	51.2^{c}	61.5^{h}	31.66
		0.3	34.7^{bc}	50.9^{c}	63.4^{g}	29.55
Control			35.7^{ab}	64.8^{a}	90.0^{a}	

DAI - Days after inoculation

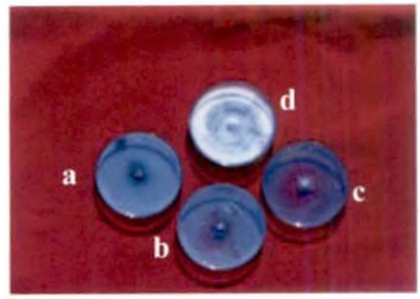
* Mean of five replications

In each column figures followed by same letter do not differ significantly according to DMRT

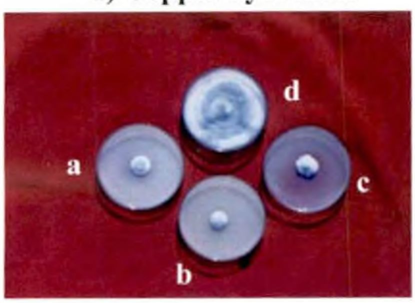
A) Fungicides



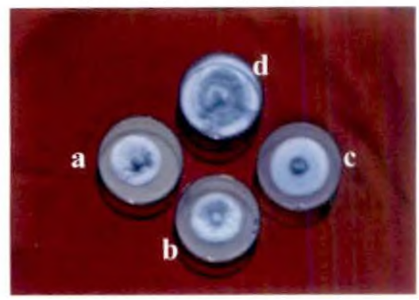
1) Copper hydroxide



2) Copper oxychloride



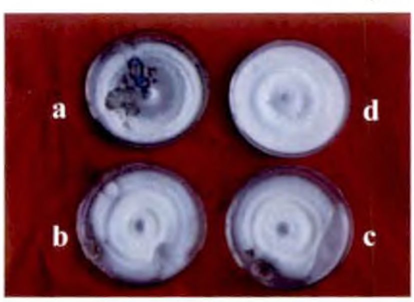
3) Captan



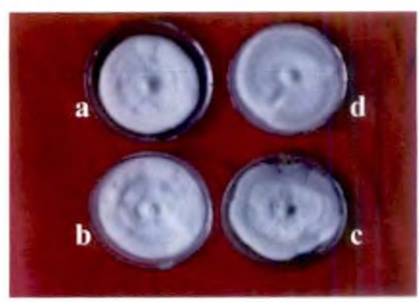
4) Mancozeb

a - 0.1 per cent concentration
b - 0.2 per cent concentration
c - 0.3 per cent concentration
d - Control

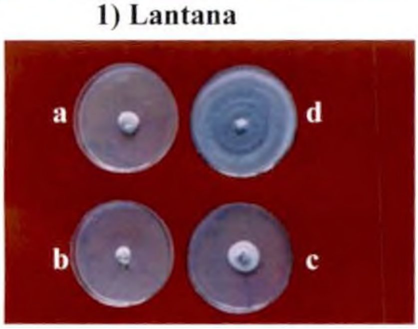
B) Botanicals



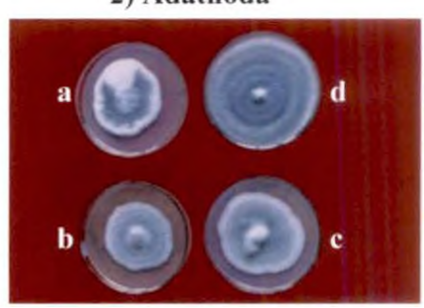
1) Lantana



2) Adathoda



3) Garlic



4) Neem oil (0.1, 0.2, 0.3 per cent)

a - 5.0 per cent concentration
b - 10.0 per cent concentration
c - 15.0 per cent concentration
d - Control

Plate 5. ***In vitro*** **evaluation of fungicides and botanicals against** ***C. gloeosporioides***

4.11.3 *In vitro* evaluation of fungicides and botanicals against *A. alternata*

4.11.3.1 Fungicides

The data on the inhibitory effect of different fungicides on *A. alternata* are given in Table 14. The data showed significant difference among the treatments on their inhibitory effect on the growth of the fungus. The control plates recorded full growth on seventh day after incubation.

Copper hydroxide, 0.2 per cent concentration recorded the maximum inhibition on growth of the pathogen (86.55 per cent) over control. It was on par with 0.1 and 0.3 per cent of copper hydroxide where the growth inhibition over control was recorded as 84.09 and 83.12 per cent respectively. It was followed by copper oxychloride at all the three concentrations and captan 0.2 per cent recorded inhibition of growth ranging from 72.66 to 76.0 per cent over control. The lowest inhibition on growth of the pathogen was noticed for 0.1 per cent mancozeb which recorded the least per cent inhibition of 49.11 over the control. (Plate 6A)

4.11.3.2 Botanicals

The analysis of data (Table 14) showed significant difference among treatments on their inhibitory effect against the fungus. Garlic at 10 per cent concentration was found most inhibitory to the growth of fungus and recorded 81.55 per cent inhibition over control. But it was on par with garlic 15 per cent concentration, recorded 79.33 per cent inhibition over control. Leaf extract of Lantana 15 per cent showed least inhibition (21.64 per cent) which was on par with 15 per cent Adathoda and 5 per cent Lantana, recorded 21.86 and 22.77 per cent inhibition respectively. All the three concentrations of neem oil showed a partial inhibition on growth of the fungus ranging from 38.44 to 42.55 per cent over control.(Plate 6B)

Effect of fungicides and botanicals on sporulation of *A. alternata* and *C. gloeosporioides* were studied and presented in Table 15. It was noted that both the organisms showed moderate sporulation with 0.2 per cent mancozeb and sparse

Table 14 *In vitro* evaluation of fungicides and botanicals against *A. alternata*

a. Fungicides

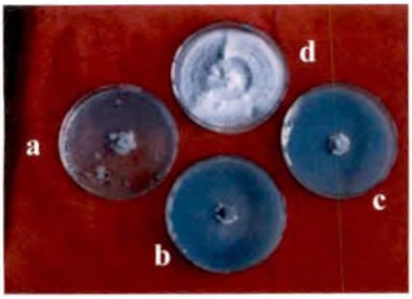
Sl. No.	Treatments	Concentration (Per cent)	DAI (diameter in mm)				Percent inhibition over control
			2nd	4th	6th	7th	
1	Copper hydroxide	0.1	9.1^{k}	9.8^{q}	12.7^{no}	14.3^{no}	84.09
		0.2	9.1^{k}	10.2^{q}	11.7^{o}	12.1^{o}	86.55
		0.3	9.9^{k}	10.8^{q}	13.8^{mno}	15.2^{n}	83.12
2	Copper oxychloride	0.1	16.1^{i}	19.7^{l}	22.2^{kl}	23.7^{kl}	73.66
		0.2	13.2^{j}	15.8^{no}	20.6^{l}	21.6^{l}	76.00
		0.3	13.2^{j}	16.5^{mn}	20.9^{l}	22.0^{l}	75.55
3	Captan	0.1	18.3^{gh}	22.2^{k}	29.8^{j}	31.6^{i}	64.89
		0.2	16.8^{hi}	18.5^{lm}	22.8^{kl}	24.6^{k}	72.66
		0.3	17.1^{ghi}	19.8^{l}	24.0^{k}	27.7^{j}	69.28
4	Mancozeb	0.1	18.8^{g}	28.6^{i}	42.7^{h}	45.8^{g}	49.11
		0.2	17.5^{ghi}	25.5^{j}	38.8^{i}	42.4^{h}	52.88
		0.3	17.9^{gh}	26.2^{j}	39.6^{i}	42.6^{h}	52.66
Control			38.1^{a}	60.8^{a}	81.8^{a}	90.0^{a}	
b. Botanicals							
1	Lantana	5	31.0^{c}	48.1^{cd}	60.6^{de}	69.5^{bc}	22.77
		10	31.2^{c}	47.2^{de}	60.1^{e}	68.0^{cd}	24.53
		15	33.5^{b}	50.3^{b}	64.5^{bc}	70.6^{b}	21.64
2	Adathoda	5	29.9^{c}	45.8^{e}	61.4^{de}	66.6^{d}	26.00
		10	27.9^{d}	49.6^{bc}	63.2^{cd}	68.0^{cd}	24.44
		15	29.6^{c}	47.3^{de}	66.3^{b}	70.4^{bc}	21.86
3	Garlic	5	12.6^{j}	16.7^{mn}	21.0^{l}	23.7^{kl}	73.66
		10	10.7^{k}	13.1^{p}	14.7^{mn}	16.6^{mn}	81.55
		15	9.8^{k}	13.9^{op}	16.2^{m}	18.6^{m}	79.33
4	Neem oil	0.1	21.1^{e}	40.9^{f}	53.2^{f}	55.4^{e}	38.44
		0.2	20.7^{f}	31.2^{h}	49.1^{g}	51.7^{f}	42.55
		0.3	21.8^{f}	34.2^{g}	49.4^{g}	53.5^{ef}	40.55
Control			38.7^{a}	59.9^{a}	81.2^{a}	90.0^{a}	

DAI - Days after inoculation

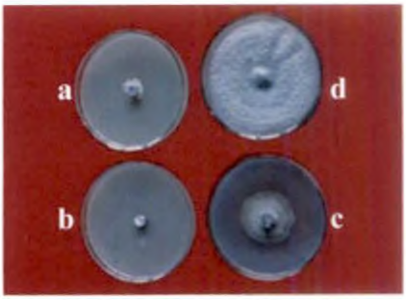
* Mean of five replications

In each column figures followed by same letter do not differ significantly according to DMRT

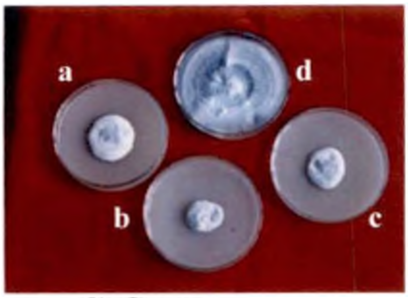
A) Fungicides



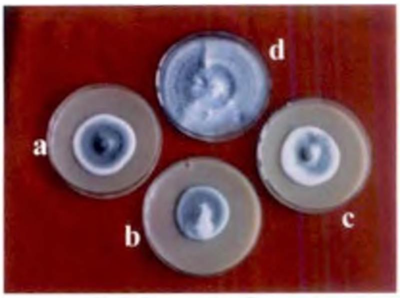
1) Copper hydroxide



2) Copper oxychloride



3) Captan



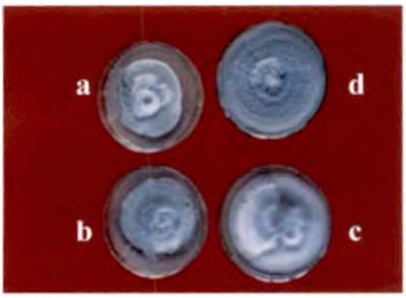
4) Mancozeb

a - 0.1 per cent concentration
b - 0.2 per cent concentration
c - 0.3 per cent concentration
d - Control

B) Botanicals

1) Lantana

2) Garlic



3) Neem oil (0.1, 0.2, 0.3 per cent)

a - 5.0 per cent concentration
b - 10.0 per cent concentration
c - 15.0 per cent concentration
d - Control

Plate 6. *In vitro* evaluation of fungicides and botanicals against *A. alternata*

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sporulation was noted with 0.1 and 0.3 per cent concentrations of mancozeb. In all concentrations of captan incorporated media sparse sporulation was noted for both pathogens, except for 0.1 per cent concentration where moderate conidial production was noticed for *C. gloeosporioides*. There was no sporulation for both fungus in media incorporated with different concentrations of copper hydroxide and copper oxychloride.

Moderate sporulation was noticed for *A. alternata* and *C. gloeosporioides* in the media incorporated with the three concentrations of Adathoda and Lantana (Table15). In 0.1 per cent concentration of neem oil incorporated media, moderate sporulation was noticed for *A. alternata* and sparse sporulation for *C. gloeosporioides*. For 0.2 and 0.3 per cent concentrations of neem oil, sporulation was absent. Also, sporulation was not noticed in the three concentrations of garlic for both the fungus.

4.11.4 *In vitro* evaluation of antagonists against leaf spot pathogens

The antagonistic activity of four fungal antagonists and one bacterial antagonist against three leaf spot pathogens were assessed as described in the materials and methods.(Plate 7)

4.11.4.1 Evaluation of fungal antagonists against C. cocciniae

The efficacy of fungal antagonists against *C. cocciniae* was assessed by dual culture method and per cent inhibition of pathogen was recorded. The results of the experiment are presented in Table 16.

The initial growth rate of the test organism and antagonist was almost same in both mono and dual culture. But the growth of antagonist was faster as compared to the pathogen except for *C. globosum*. On the third day of incubation growth of *T. viride* was 49.4 mm against 29.8 mm of pathogen, *T. harzianum* showed 48.7 mm against 30.3 mm of pathogen and *A. niger* showed 46.4 mm against 30.7 mm of pathogen. On four days after incubation, *T. viride, T. harzianum* and *A. niger* started over growing pathogen. *C. globosum* recorded

Table 15a Effect of fungicides on sporulation of leaf spot pathogens

Sl. No.	Treatments	Concentration (Per cent)	*A. alternata*	*C. gloeosporioides*
1	Copper hydroxide	0.1	-	-
		0.2	-	-
		0.3	-	-
2	Copper oxychloride	0.1	-	-
		0.2	-	-
		0.3	-	-
3	Captan	0.1	+	++
		0.2	+	+
		0.3	+	+
4	Mancozeb	0.1	++	++
		0.2	+	+
		0.3	+	+

15b Effect of botanicals on sporulation of leaf spot pathogens

Sl. No.	Treatments	Concentration (Per cent)	*A. alternata*	*C. gloeosporioides*
1	Adathoda	5	++	++
		10	++	++
		15	++	++
2	Lantana	5	++	++
		10	++	++
		15	++	++
3	Garlic	5	-	-
		10	-	-
		15	-	-
4	Neem oil	0.1	++	+
		0.2	-	-
		0.3	-	-

+++ : Good sporulation
++ : Moderate sporulation
+ : Sparse sporulation
- : Absent / no sporulation

Table 16. *In vitro* evaluation of antagonists against *C. cocciniae*

Antagonists used		Days after incubation (diameter mm)																Antagonism index	Type of Antagonism
		1		2		3		4		5		6		7		8			
		T	A	T	A	T	A	T	A	T	A	T	A	T	A	T	A		
T.viride	D	15.1	24.2	20.3	36.2	29.8	49.4	19.6	60.6	11.1	74.2	0	90						II
	M	15.4	26.4	22.8	40.8	33.8	55.3	44.8	71.1	56.9	90	68.2	90	79.8	90	90	90	1500	
T.harzianum	D	14.9	22.6	21.0	36.1	30.3	48.7	20.9	60.1	11.4	73.7	0	90					1500	II
	M	15.4	24.5	22.8	38.6	33.8	54.6	44.8	70.8	56.9	90	68.2	90	79.8	90	90	90		
C. globosum	D	15.4	19.6	21.2	28.4	32.2	39.7	43.3	47.9	36.0	54.0	26.1	63.8	14.2	75.3	2.5	87.5	1166.	I
	M	15.4	21.1	22.8	30.7	33.8	41.9	44.8	49.6	56.9	57.7	68.2	69.4	79.8	80.2	90	90	4	
A. niger	D	15.0	26.1	20.8	37.4	30.7	46.4	26.6	59.1	19.1	68.5	10.0	79.2	0	90			1200	II
	M	15.4	31.8	22.8	39.2	33.8	52.8	44.8	64.3	56.9	76.1	68.2	90	79.8	90	90	90		

T - Test organism
A - Antagonist
D - Dual culture
M - Mono culture

I - Homogenous
II - Overgrowth
III - Cessation of growth
IV- Aversion

47.9 mm growth on the fourth day against 43.3 mm growth of pathogen. After the fourth day, over growth by antagonist was noticed and a free intermingling of hyphae between pathogen and antagonist was noticed. For *T. viride* and *T. harzianum* over growth was completed by six days after incubation and for *A. niger* it was completed by seven days after incubation. Almost over growth was noticed for *C. globosum* on the eight days after incubation, which measured 87.5 mm for antagonist and 2.5 mm for pathogen. The mechanism of antagonism noticed in *T. viride, T. harzianum* and *A. niger* was over growth on the pathogen.

4.11.4.2 Evaluation of fungal antagonists against C. gloeosporioides

The growth rate of test organism and antagonist was almost same initially. Rate of growth of antagonist was faster compared to that of pathogen as evidenced after three days of incubation (Table 17). The measurement of *T. viride* showed 51.8 mm against 39.0 mm of pathogen, *T. harzianum* showed 50.1mm against 39.8 mm of pathogen and *A. niger* showed 50.0 mm against 39.9 mm of pathogen. Over growth on pathogen started four days after incubation for these antagonists. *C. globosum*, on four days after incubation, showed 44.1 mm growth against 44.6 mm of pathogen and on the fifth day onwards slight over growth started. Between the pathogen and *C. globosum*, there was a slight aversion in growth and no complete over growth was noticed. On seventh day after incubation, *C. globosum* measured 53.2 mm against 36.8 mm of pathogen. *T. viride* and *T. harzianum* showed complete over growth on 6 days after incubation. For *A. niger,* almost complete over growth was noticed and measured 80.5 mm against 9.4 mm of pathogen.

4.11.4.3 Evaluation of fungal antagonists against A. alternata

The initial growth rate for antagonist and pathogen was same, but growth rate of antagonist became faster compared to the pathogen on the third day onwards (Table 18). On the third day, *T. viride* measured 50.4 mm diameter against 39.2 mm of pathogen and *T. harzianum* showed 50.1 mm against 39.8 mm of pathogen. Both the organism showed over growth on fourth day onwards and completed on six days after incubation. *A. niger* showed 47 2 mm growth on third

Table 17. *In vitro* evaluation of antagonists against *C. gloeosporioides*

Antagonists used		Days after incubation (Diameter in mm)																Antagonism index	Type of Antagonism
		1		2		3		4		5		6		7		8			
		T	A	T	A	T	A	T	A	T	A	T	A	T	A	T	A		
T. viride	D	30.2	28.4	36.8	38.6	39.0	51.8	23.8	66.1	8.4	81.4	0	90					1500	II
	M	31.6	28.8	43.5	40.1	56.7	55.4	67	70.7	79	90	90	90						
T. harzianum	D	31.0	27.6	40.4	37.8	39.8	50.1	26.1	63.6	10.5	79.5	0	90					1500	II
	M	31.6	27.1	43.5	39.2	56.7	53.4	67	69.8	78	90	90	90						
C. globosum	D	31.3	21.3	40.3	32.2	44.4	40.6	44.6	44.1	41.2	48.6	38.6	51.3	36.8	53.2			1166.4	IV& II
	M	31.6	21.5	43.5	32.8	56.7	41.4	67	48.2	78	56.7	90	67.5	90	78.6	90	90		
A. niger	D	26.0	31.9	30.4	44.7	39.9	50.0	22.5	67.3	17.9	72	9.4	80.5					1200	II
	M	31.6	32.3	43.5	44.8	56.7	54.9	67	68	78	76.1	90	90						

T - Test organism
A - Antagonist
D - Dual culture
M - Mono culture

I- Homogenous
II - Overgrowth
III - Cessation of growth
IV - Aversion

day, against 42.6 mm growth of pathogen and over growth started on fourth day onwards. Here, the over growth by the antagonist was not completed and measured 81.6 mm for *A. niger* and 8.4 mm for pathogen after seven days of incubation. For *C. globosum*, on the fourth day of growth showed 47 mm growth against 44 mm growth of pathogen. Over growth started on fifth day onwards and a slight aversion between the hyphae of pathogen and antagonist was noticed. On the seventh day after incubation antagonist measured 53.8 mm as against 36.2 mm of pathogen.

Among the four fungal antagonists tested, *T. viride* and *T. harzianum* were found to be the most effective recording the highest AI value of 1500 against all the three pathogens followed by *A. niger*. It is also observed from the table that eventhough *C. globosum* was least inhibitory to *C. gloeosporiodes* and *A. alternata,* it was effective against *C. cocciniae* as evident from the AI value (1166.4) recorded.

4.11.5 In vitro evaluation of *P. fluorescens* against leaf spot pathogens

Among the three methods adopted (ring method, square method and streak method) ring method was found most effective in testing the antagonistic property of *P. fluorescens* against the different pathogen.

From Table 19, it is found that *P. fluorescens* recorded 48.43 per cent inhibition of growth of *C. cocciniae* over control where as in *C. gloeosporioides* and *A. alternata* it was 62.78 and 59.50 per cent respectively. The inhibition zone of 2mm and 1mm was developed between the antagonist and test organism as evidenced in case of *A. alternata* and *C. gloeosporioides* .

4.12 Evaluation on phytotoxic effect of copper fungicides on ivy gourd

Phytotoxic effect of copper fungicides used in the study i.e., copper oxychloride (Fytolan) and copper hydroxide (Kocide) on ivy gourd leaves were tested. Copper oxychloride at the three concentration (0.1, 0.2 and 0.3 per cent) showed no adverse effect on plants. Copper hydroxide at 0.1 and 0.2 per cent concentrations also not showed any phytotoxic effect, but at 0.3 per cent

Table 18. *In vitro* evaluation of antagonists against *A. alternata*

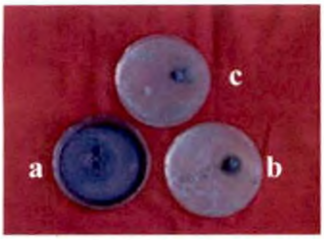
Antagonists used		Days after incubation (diameter in mm)																Antagonism index	Type of Antagonism
		1		2		3		4		5		6		7		8			
		T	A	T	A	T	A	T	A	T	A	T	A	T	A	T	A		
T. viride	D	24.1	26.3	35.6	39.8	39.2	50.4	20.8	69.2	9.1	80.7	0	90					1500	II
	M	24.7	26.8	39.6	40.4	48.9	56.6	59.8	71.2	70.1	90	81.3	90	90	90				
T. harzianum	D	24.4	23.7	36.1	38.4	39.8	50.1	24.2	65.3	13.6	76.4	0	90					1500	II
	M	24.7	23.8	39.6	39.7	48.9	54.7	59.8	70.5	70.1	90	81.3	90	90	90				
C. globosum	D	24.0	19.7	35	31.5	41.0	41.0	44.0	47.0	40.7	49.2	38.4	51.4	36.2	53.8			538.2	IV & II
	M	24.7	20.0	39.6	31.9	48.9	42.4	59.8	49.1	70.1	56.5	81.3	68.3	90	79.9	90	90		
A.niger	D	23.9	31.4	34.6	38.0	42.6	47.2	31.8	58.1	19.5	70.3	10.2	78.6	8.4	81.6			1088.4	II
	M	24.7	32.6	39.6	39.1	48.9	50.2	59.8	62.3	70.1	76.8	81.3	90	90	90				

T - Test organism
A - Antagonist
D - Dual culture
M - Mono culture

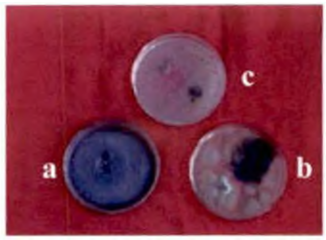
I - Homogenous
II- Overgrowth
III - Cessation of growth
IV - Aversion

Table 19. *In vitro* evaluation of *P. fluorescens* against different leaf spot pathogens

Leaf spot pathogens	Days after incubation (mm)								Percent inhibition (PI)
	1		2		3		4		
	T	A	T	A	T	A	T	A	
C.cocciniae	10.8	6.2	15.3	8.5	19.4	10.0	23.1	10.0	48.43
C. gloeosporioides	14.3	6.3	18.6	8.4	21.1	9.8	-	-	62.78
A. alternata	12.2	6.2	16.7	8.4	19.8	9.7	-	-	59.50

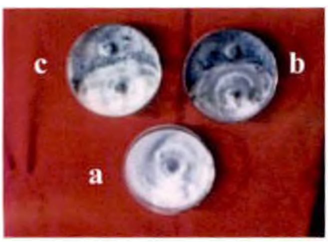


a - *C. cocciniae*
b - *T. viride*
c - *T. harzianum*



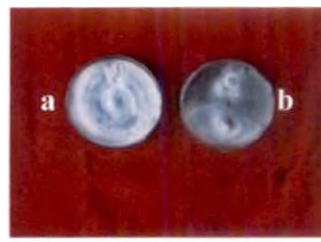
a - *C. cocciniae*
b - *C. globosum*
c - *A. niger*

a - *C. cocciniae*
b - *P. flourescens*

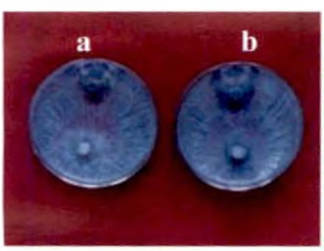


a - *C. gloeosporioides*
b - *T. viride*
c - *T. harzianum*

a - *C. gloeosporioides*
b - *A. niger*

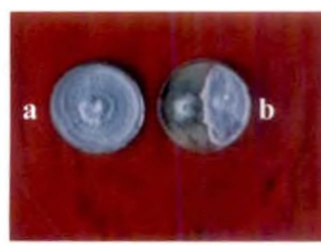


a - *C. gloeosporioides*
b - *C. globosum*



a - *T. viride*
b - *T. harzianum*

a - *A. alternata*
b - *A. niger*



a - *A. alternata*
b - *C. globosum*

a - *A. alternata*
b - *P. flourescens*

Plate 7. *In vitro* evaluation of antagonistic organisms against different leaf spot pathogens

concentration, the middle and upper leaves showed marginal yellowing and drying. Finally those leaves became brown coloured and dried.

4.13 Pot culture studies on the evaluation of various fungicides, botanicals and antagonists against leaf spot diseases of ivy gourd

Effect of different fungicides, botanicals and antagonistic organisms on the management of leaf spot diseases of ivy gourd was studied by pot culture experiment. Three sprayings were given on naturally infected ivy gourd plants at an interval of 15 days after the first appearance of the leaf spot disease. Observations on the disease incidence and severity were recorded just before the first spraying and also on every tenth and fourteenth day of each spraying. The concentration of fungicides (0.2per cent) and botanicals (10per cent) which recorded minimum inhibitory concentration values (MIC) and efficient fungal and bacterial antagonists from the *in vitro* evaluation were selected for the pot culture studies.

From the data presented in the Table 20, it was observed that there was significant difference among the treatments on per cent disease severity which was recorded before and after each spraying. Per cent disease incidence was calculated before and after each spraying which was found to be cent per cent throughout the experiment.

Observations on disease severity on ten days after spraying revealed that the plants in treatment T_2 (copper oxy chloride) recorded the minimum disease severity of 21.30 which was on par with T_8 (garlic) which showed 22.7 per cent disease severity. The treatments T_4(mancozeb) and T_5(Adathoda) showed the maximum disease severity of 37.0 and 37.3 per cent respectively. It was on par with control (T_{12}) and T_1, T_3, T_6, T_7 and T_{11}.

On fourteen days after spraying the treatments T_2 (copper oxychloride) recorded the minimum per cent disease severity of 20.0 per cent and maximum per cent disease reduction of 39.4 per cent over control. It was on par with T_8 (garlic) and T_9 (*T. viride*) showed the per cent disease severity of 21.3 and 23.0 and per cent reduction of 35.4 and 30.3 respectively. The highest PDS (38.0 per cent) was recorded by T_4 (mancozeb) which showed a per cent increase of 15.1 per cent over control. The treatment T_4 was on par with T_3, T_5, T_7 and T_{12} (control).

The analysis of the data recorded ten days after the second spraying revealed significant difference among the treatments. Copper oxychloride at 0.2 per cent concentration (T_2) recorded the minimum disease severity of 18.7 per cent. It was on par with all the treatments, except T_4, T_5, T_7 and T_{12}.The highest disease severity was recorded by mancozeb (T_4) (38.7 per cent).

The data on disease severity on fourteen days after spraying of second spray showed that copper oxychloride (0.2 per cent) concentration was the best among the treatments to control the leaf spot diseases of ivy gourd. It recorded 17.7 per cent disease severity and 47.5 per cent reduction of diseases over control. It was found to be on par with T_1, T_6, T_8, T_9, T_{10} and T_{11}. The treatment T_4 (mancozeb) recorded the maximum PDS of 36.3 per cent and 7.9 per cent increase in per cent disease severity over control.

Observations of tenth day after the third spraying recorded significant difference among the treatments. The treatment T_2 (copper oxychloride) recorded the lowest disease severity (12.7 per cent) and was on par with T_9 and T_{10} recorded disease severity of 13.7 and 19.7 per cent respectively. It was also on par with T_1 (copper hydroxide) and T_8 (garlic). The treatment T_4 (mancozeb) recorded the highest per cent disease severity of 31.7 which was on par with T_3, T_5, T_7 and T_{12}.

The data recorded on fourteenth day after the third spraying also showed significance among the treatments. Copper oxychloride (T_2) was recorded as the most efficient fungicide to control the leaf spot pathogens. It recorded 8.0 per cent disease severity and 75.0 per cent reduction of disease over control. It was followed by T_9 (garlic) which recorded 8.3 per cent disease severity and 73.9 per cent reduction over control. These treatments were on par with T_1, T_6, T_8 and T_{10}. Control plants (T_{12}) recorded the maximum per cent disease severity (32.0 per cent) and it was on par with T_4 (mancozeb)and T_7 (Lantana)(Fig.7).

Table20 Effect of fungicides, botanicals and antagonists on per cent disease severity under pot culture studies

Treatments	Per cent disease severity									
	Before first spraying	After first spraying			After second spraying			After third spraying		
		10 DAS	14 DAS	Per cent reduction over control	10 DAS	14 DAS	Per cent reduction over control	10 DAS	14 DAS	Per cent reduction over control
T_1	30.7^{ab}	26.7^{abc}	25.7^{cd}	22.2	20.0^{cd}	19.0^{de}	43.6	14.7^{de}	9.3^{ef}	70.8
T_2	20.0^{c}	21.3^{c}	20.0^{d}	39.4	18.7^{d}	17.7^{e}	47.5	12.7^{e}	8.0^{f}	75.0
T_3	29.3^{abc}	34.7^{ab}	33.7^{abc}	-2.0	30.7^{abcd}	29.0^{abcd}	13.9	24.7^{abc}	19.0^{cd}	40.6
T_4	34.0^{a}	37.0^{a}	38.0^{a}	-15.1	38.7^{a}	36.3^{a}	-7.9	31.7^{a}	29.0^{ab}	9.4
T_5	24.7^{abc}	37.3^{a}	36.7^{ab}	-11.1	34.7^{ab}	33.7^{ab}	0	28.0^{abc}	23.7^{bc}	26.0
T_6	22.7^{bc}	27.0^{abc}	27.3^{bcd}	17.2	26.3^{abcd}	25.3^{bcde}	24.8	22.7^{bcd}	15.7^{def}	67.0
T_7	25.7^{abc}	33.7^{ab}	33.7^{abc}	2.0	31.7^{abc}	31.3^{abc}	6.9	30.0^{ab}	26.0^{abc}	18.8
T_8	22.7^{bc}	22.7^{c}	21.3^{d}	35.4	20.0^{cd}	19.0^{de}	43.6	15.3^{de}	9.0^{ef}	71.9
T_9	25.3^{abc}	24.3^{bc}	23.0^{d}	30.3	20.3^{cd}	19.0^{de}	43.6	13.7^{e}	8.3^{ef}	73.9
T_{10}	24.0^{bc}	26.0^{bc}	25.0^{cd}	24.2	24.0^{bcd}	23.0^{cde}	31.7	19.7^{e}	15.0^{def}	53.1
T_{11}	28.0^{abc}	29.3^{abc}	28.3^{bcd}	15.15	27.0^{abcd}	25.3^{bcde}	24.8	22.7^{bcd}	16.0^{de}	50.0
T_{12}	27.3^{abc}	31.0^{abc}	33.0^{abc}	-	34.3^{ab}	33.7^{ab}	0	31.0^{ab}	32.0^{a}	0

T_1 - Copper hydroxide
T_2 - Copper oxychloride
T_3 - Captan
T_4 - Mancozeb
T_5 - Adathoda
T_6 - Neem oil
T_7 - Lantana
T_8 - Garlic
T_9 – *T. viride*
T_{10} – *T. harzianum*
T_{11} – *P. fluorescens*
T_{12} - Control

In each column figures followed by same letter do not differ significantly according to DMRT
DAS - Days after spraying

4.14 Field evaluation on the efficacy of selected fungicides, botanicals and antagonists against leaf spot diseases of ivy gourd.

Effectiveness of selected fungicides, botanicals and antagonists in reducing the severity of different leaf spot diseases under natural conditions were studied through a field experiment laid out at College of Horticulture, Vellanikkara. The treatments which were found effective against all leaf spot organisms in pot culture experiment were used for the field evaluation.

Appearance of disease was noticed 60 days after planting. Spraying with different treatments and assessment of per cent disease incidence, per cent disease severity and per cent inhibition over control were done. The data of the experiment are presented in Table 22.

The plants under various treatments recorded cent per cent disease incidence throughout the period of this study. The observations recorded 10 days after the first spraying showed significant difference among treatments, on their effect to control the leaf spot diseases. It was noticed that in all the treatments except control (T_7), the per cent disease severity was decreased compared to that before spraying. The lowest PDS was observed in plants sprayed with garlic (T_4, 15.9 per cent). It was on par with T_2 (copper oxychloride) which recorded 16.9 per cent disease severity. The highest PDS was noticed in T_7 (26.0 per cent).

The analysis of the observations recorded on 14 days after the first spraying showed significant difference among the treatments. The treatment T_2 recorded the lowest PDS of 16.1 per cent which was on par with T_4 (17.0 per cent). The highest per cent reduction over control was noticed in T_2 (39.5 per cent). It was followed by T_4 which showed 36.0 per cent reduction. The control plants (T_7) recorded the highest PDS (26.6 per cent).

From the data recorded on 10 days after second spraying, it was found that the treatments differ significantly each other in their efficiency to control the leaf spot diseases. The treatment T_2 (copper oxychloride) recorded the lowest PDS

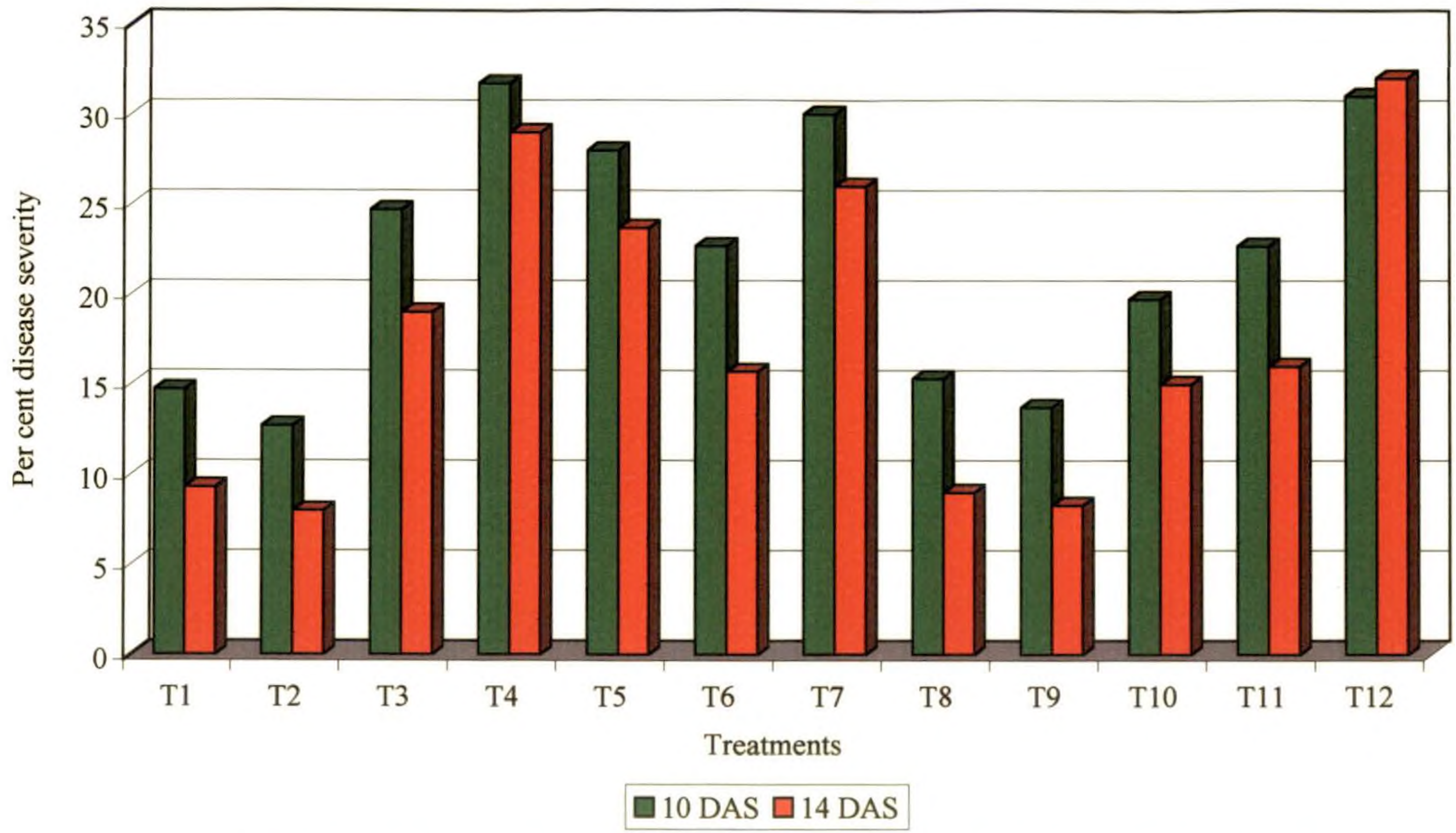


Fig. 7. Effect of different treatments on per cent disease severity of leaf spot diseases after final spraying during pot culture experiment

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The analysis of the observations recorded on 14 days after the first spraying showed significant difference among the treatments. The treatment T_2 recorded the lowest PDS of 16.1 per cent which was on par with T_4 (17.0 per cent). The highest per cent reduction over control was noticed in T_2 (39.5 per cent). It was followed by T_4 which showed 36.0 per cent reduction. The control plants (T_7) recorded the highest PDS (26.6 per cent).

From the data recorded on 10 days after second spraying, it was found that the treatments differ significantly each other in their efficiency to control the leaf spot diseases. The treatment T_2 (copper oxychloride) recorded the lowest PDS

Table 21 Field evaluation of selected fungicides, botanicals and antagonists against leaf spot diseases of ivy gourd

Treatments	Per cent disease severity									
	Before first spraying	After first spraying			After second spraying			After third spraying		
		10 DAS	14 DAS	Per cent reduction over control	10 DAS	14 DAS	Per cent reduction over control	10 DAS	14 DAS	Per cent reduction over control
T$_1$	27.0^{a}	23.9^{b}	22.0^{b}	17.3	19.3^{bc}	16.0^{de}	43.7	13.0^{c}	10.0^{c}	65.0
T$_2$	19.5^{e}	16.9^{d}	16.1^{d}	39.5	14.1^{d}	13.4^{f}	52.8	11.5^{d}	9.5^{c}	66.8
T$_3$	23.3^{c}	21.0^{c}	19.6^{c}	26.3	18.4^{c}	18.0^{c}	36.6	16.8^{b}	14.7^{b}	48.6
T$_4$	21.8^{d}	15.9^{d}	17.0^{d}	36.0	15.3^{d}	15.0^{e}	47.2	12.7^{c}	9.4^{c}	67.1
T$_5$	22.1^{d}	20.1^{c}	20.2^{c}	24.1	18.0^{c}	16.3^{d}	42.6	12.7^{c}	9.7^{c}	66.0
T$_6$	25.1^{b}	23.2^{b}	22.1^{b}	16.9	20.0^{b}	19.6^{b}	31.0	16.0^{b}	14.5^{b}	49.3
T$_7$	24.6^{b}	26.0^{a}	26.6^{a}	-	27.0^{a}	28.4^{a}	-	27.4^{a}	28.6^{a}	-

T$_1$ - Copper hydroxide
T$_2$ - Copper oxychloride
T$_3$ - Neem oil
T$_4$ - Garlic
T$_5$ - *Trichoderma viride*
T$_6$ - *Pseudomonas fluorescens*
T$_7$ - Control

In each column figures followed by same letter do not differ significantly according to DMRT

DAS - Days after spraying

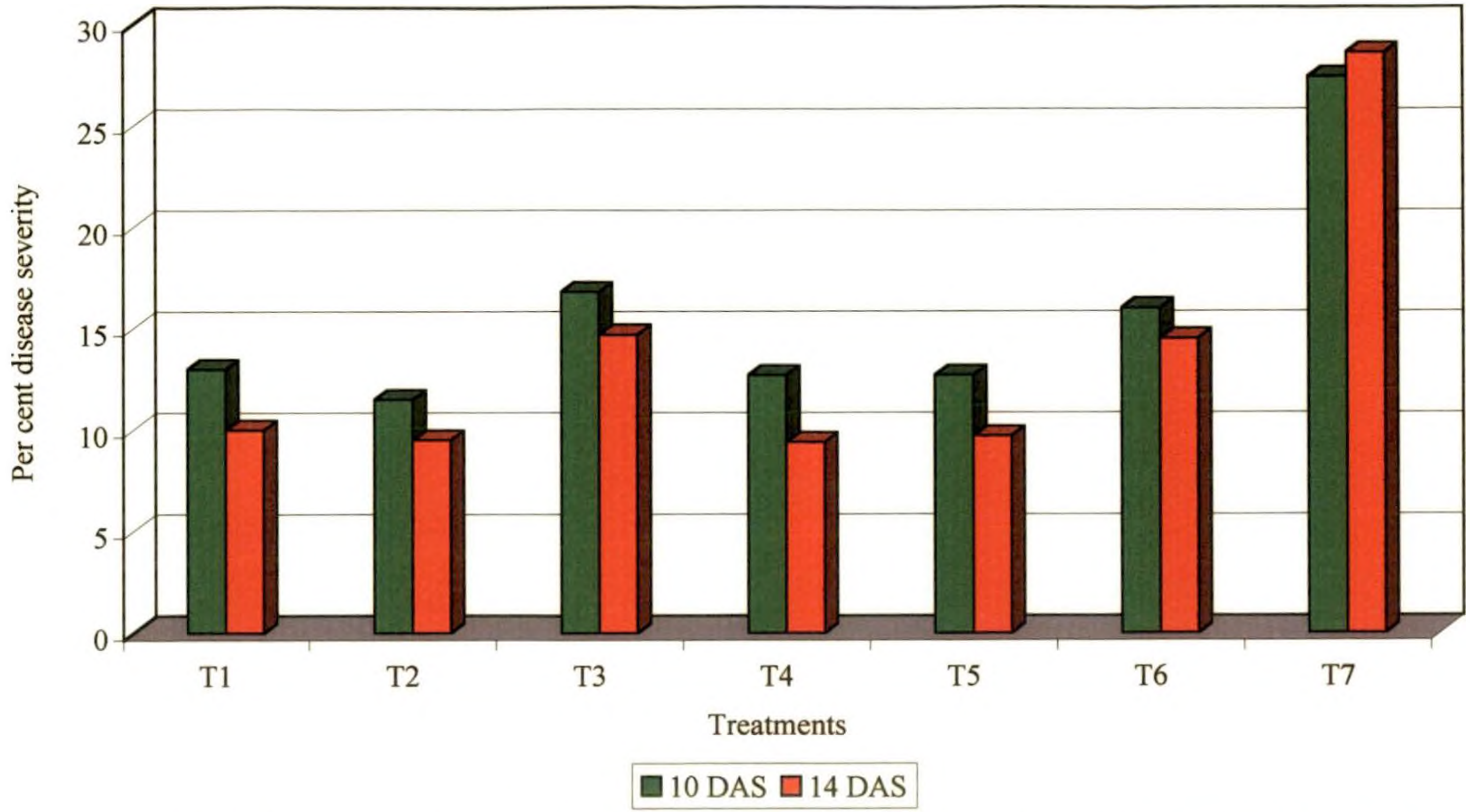

Fig. 8. Effect of different treatments on per cent disease severity of leaf spot diseases after final spraying during field experiment

(14.1) which was on par with T_4 (garlic) showing 15.3 per cent disease severity. The control (T_7) recorded highest PDS of 27.0 per cent.

The second observation after the second spraying, taken on fourteen days after spraying also showed significant difference among the treatments. The treatment T_2 recorded the lowest PDS (13.4) and highest per cent reduction of 52.8 over the control. It was followed by T_4, recorded PDS of 15.0 per cent and 47.2 per cent reduction over control. The highest PDS was showed by the plants without any treatment (T_7, 28.4 per cent).

After the third spray, the observations on PDS was recorded on 10 DAS, which also showed significant difference between the treatments. The treatment T_2 (copper oxychloride, 0.2 per cent) recorded the lowest PDS (11.5).It was followed by T_1, T_4 and T_5 which were on par with each other. The treatment T_7 (control) recorded the maximum PDS (27.4).

On 14 days after the third spraying, the lowest PDS (9.4) and highest per cent reduction over control (67.1) were recorded by T_4 (garlic), and it was on par with T_1, T_2 and T_5 which showed PDS of 10.0, 9.5 and 9.7 respectively. The highest PDS was recorded by the control plants (28.6).(Fig 8)

4.14.1 Field evaluation of fungicides, botanicals and antagonists against fruit spot in ivy gourd

Effectiveness of selected fungicides, botanicals and antagonists in controlling the incidence and severity of fruit spot of ivy gourd under natural condition were also evaluated during field experiment. The data is presented in Table 22.

The observations on incidence and severity of fruit spot taken from plants grown under different treatments showed significant difference among them. After the first spraying the lowest disease incidence (8.3 per cent) and severity (8.0 per cent) were noticed for T_4 (garlic) and T_5 (*T. viride*). Except T_7 (control), all treatments were found on par in per cent disease severity.

Table22 Field evaluation of selected fungicides, botanicals and antagonists against fruit spot on ivy gourd

Treatments	Before spraying		First spraying		Second spraying		Third spraying		Yield/plant (g)
	PDI	PDS	PDI	PDS	PDI	PDS	PDI	PDS	
T_1	25 (0.524)^{ab}	12 (3.431)^{bc}	11.1 (0.335)^{bc}	8 (2.828)^{b}	8.3 (0.292)^{b}	4 (2.121)^{b}	0 (0.000)^{c}	0 (0.707)^{d}	570^{a}
T_2	16.7 (0.400)^{b}	8 (2.667)^{c}	16.7 (0.421)^{b}	12 (3.431)^{b}	0 (0.000)^{c}	0 (0.707)^{c}	0 (0.000)^{c}	0 (0.707)^{d}	583.3^{a}
T_3	16.7 (0.421)^{ab}	8 (2.828)^{c}	11.1 (0.335)^{bc}	12 (3.431)^{b}	8.3 (0.292)^{b}	8 (2.915)^{b}	8.3 (0.292)^{b}	4 (2.121)^{c}	530^{a}
T_4	11.1 (0.335)^{b}	10.6 (3.219)^{bc}	8.3 (0.292)^{c}	8 (2.667)^{b}	0 (0.000)^{c}	0 (0.707)^{c}	0 (0.000)^{c}	0 (0.707)^{d}	576.6^{a}
T_5	11.1 (0.335)^{b}	12 (3.431)^{bc}	8.3 (0.292)^{c}	8 (2.828)^{b}	8.3 (0.292)^{b}	4 (2.121)^{b}	0 (0.000)^{c}	0 (0.707)^{d}	581.6a
T_6	33.3 (0.613)^{a}	20 (4.457)^{ab}	11.1 (0.335)^{bc}	12 (3.431)^{b}	11.1 (0.335)^{b}	8 (2.768)^{b}	8.3 (0.292)^{b}	4 (2.121)^{c}	560^{a}
T_7	33.3 (0.611)^{a}	32 (5.627)^{a}	33.3 (0.613)^{a}	28 (5.282)^{a}	33.3 (0.611)^{a}	32 (5.694)^{a}	33.3 (0.615)^{a}	34 (5.872)^{b}	306.6^{b}

T_1 - Copper hydroxide
T_2 - Copper oxychloride
T_3 - Neem oil
T_4 - Garlic
T_5 - *Trichoderma viride*
T_6 - *Pseudomonas fluorescens*
T_7 - Control

In each column figures followed by same letter do not differ significantly according to DMRT
Data given in parenthesis are transformed values
PDI - Per cent disease incidence.
PDS - Per cent disease severity

In treatments, T_2 (copper oxychloride) and T_4 (garlic) no disease incidence was recorded after the second spraying. The maximum incidence (33.3) and severity (32.0) was observed in control. The treatments T_1, T_3, T_5 and T_6 recorded disease severity ranging only from 4 to 8 per cent.

After the third spray, the disease incidence and disease severity for treatments T_1, T_2, T_4 and T_5 (copper oxychloride, copper hydroxide, garlic and *T.viride*) recorded as zero. Highest per cent disease incidence (33.3) and maximum per cent disease severity (34 per cent) were observed for T_7 (control). The treatments T_3 (neem oil)and T_6 (*P.fluorescens*) were found on par and recorded 8.3 per cent disease incidence and 4 per cent disease severity for both the treatments.

In the case of yield, plants sprayed with all treatments gave a higher yield when compared to the control plots which recorded only 306.6 g/plant. Among the treatments, T_2 (copper oxychloride) recorded highest yield of 583.3 g/plant and was found on par with T_1, T_3, T_4, T_5 and T_6.

Discussion

5. DISCUSSION

Ivy gourd is an important cucurbitaceous vegetable grown in almost all parts of India. The crop is affected by various diseases of which leaf spots caused by *Colletotrichum gloeosporioides*(Penz.)Sacc., *Cercospora cocciniae* Munjal, Lall and Chona and *Alternaria alternata* (Fr.) Keissler are the most important. The first report on leaf spot of ivy gourd caused by *Cercospora* sp. was given by Jain (1950) from New Delhi. There were reports on leaf and fruit infection caused by the above mentioned organisms from various parts of India. But a search on literature did not reveal any information on research conducted on ivy gourd from Kerala. In view of the above facts, an investigation was carried out to study various aspects of leaf spot diseases of ivy gourd particularly the etiology, host resistance, disease management and influence of climatic factors which will add to our knowledge on the diseases.

At first, a survey was conducted in vegetable field of Department of Olericulture, College of Horticulture, Vellanikkara and farmer's field at Elanad and Cherumkuzhi in Trichur district to study the occurrence of leaf spot diseases and for the collection of diseased samples. Three different types of leaf spots were noticed and isolation of pathogens from these various spots yielded different fungal growth in the media. Isolation of pathogens from the different types of spots on different media showed the association of three fungi viz. *Cercospora* sp, *Colletotrichum* sp, and *Alternaria* sp.

The pathogenicity of these organisms was proved by artificial inoculation on ivy gourd plants under *in vitro* and *in vivo* conditions. The symptoms produced under artificial conditions were almost similar to those observed under natural condition. The same pathogens causing leaf spot diseases were reported on ivy gourd from different parts of India except from Kerala by various workers. Jain (1950), Munjal *et al.* (1959), Rangaswami and Chandrasekaran (1961) and Rao (1962) reported *C. cocciniae* on ivy gourd as leaf spot pathogen from various places.

Symptomatological study is very helpful for the correct identification of the disease and the causal agent and there by help to adopt effective management practice. Symptom expression in diseased plants occur at the later stage of infection chain and is the most important part in the chain of events. Therefore, symptomatology of the various leaf spot diseases occurred at the different locations were studied in detail.

In the present study, it was found that the symptoms produced by the same pathogen in all the three locations were almost same. *Cercospora* sp produced white necrotic spots with yellow halo and water soaked areas on lower surface of the leaves. Later, the central area of the spots turned dry and papery. Black dots of conidiophores were always noticed at the centre of spots. Symptoms were observed on middle aged leaves and 20-25 spots per leaf of about 1-2mm diameter were noticed during severe infection. Jain (1950) also described same type of symptoms on ivy gourd. In the case of leaf spots caused by *Colletotrichum* sp, initially a water soaked spot developed, which later turned brown, circular necrotic lesions with brown margin and yellow halo, having 1-2.5mm diameter and infection mainly noticed on middle aged leaves. During serious infection, some times 25-30 spots were noticed. In this case also black dots were seen at the centre of spots which were the acervuli of the pathogen. One of the characteristic symptoms produced by *Colletotrichum* sp was the shot hole symptom which was noticed on the infected leaves of ivy gourd. Fruits were found infected by *Colletotrichum* sp which produced circular sunken reddish brown spots. Two different species of *Colletotrichum*, viz., *C. lagenarium* and *C. gloeosporioides* were reported from the fruits of ivy gourd by Rao (1964) and Laxminaranyana and Reddy (1976) respectively. The symptoms described on fruits by these workers are similar to that noticed in the present study. But there is no report on leaf spot disease caused by *C. gloeosporioides* on ivy gourd. During the survey it was also noted that combined infection of *Colletotrichum* sp and *Cercospora* sp was more prevalent in all the three locations. The spots appeared as circular reddish brown on the upper surface and water soaked area on the lower surface of leaves. On such spots conidiophores of *Cercospora* sp and acervuli of *Colletotrichum* sp were also noticed which appeared as black dots on the centre of the spots. The search of literature did not give any information about such combined infection of *Cercospora* sp and *Colletotrichum* sp on ivy gourd.

The leaf spot incited by *Alternaria* sp initiated as water soaked lesion and later turned to brown large irregular lesion with concentric zonations with clear yellow halo. Here, larger lesions of size 3-5mm in diameter was observed on older leaves.The same symptom was observed by Bhargava and Singh (1985) for *Alternaria* infection in watermelon. Symptoms observed on artificial inoculation were almost same as those produced under natural condition for all the pathogens.

The cultural and morphological characters of the pathogens were studied for the proper identification of the organism by using PDA as well as selective media. Both Carrot Leaf Extract Agar and Coccinia Leaf Extract Agar media were used for the study of *Cercospora* sp and *Colletotrichum* sp, and *Alternaria* sp were studied in PDA medium. Carrot leaf extract agar has been reported as good medium for growth and sporulation of some species of *Cercospora* (Kilpatrick and Johnson, 1956; Rangaswami and Chandrasekaran, 1962). The cultural characters of the organism on Carrot Leaf Extract Agar observed during this study were found almost similar to that described by the earlier workers. The characters showed by the organisms on Carrot Leaf Extract Agar and Coccinia Leaf Extract Agar media were found almost same. This organism showed slow growth compared to the other two pathogens. It took eight days to cover the 9 cm diameter growth in Petridish, where as *Colletotrichum sp* and *Alternaria sp* showed a slight faster growth and attained 9cm diameter growth in six and seven days respectively. The colony of *Cercospora sp* showed a light pink colour with black dots on the aerial part of the growth. Anastomosis between the matured hyphae was found to be a common phenomenon in the culture of *Cercospora sp*. Here, two hyphae grow together and by the hyphal out growth they connected together, which appeared as a ladder like structure. The same phenomenon was also observed by Jain (1950) in *Cercospora* sp isolated from ivy gourd. The conidial characters and measurements were taken from the spores collected from the naturally infected leaves, because sporulation was absent in the culture. The other morphological characters observed were also same as those described by Jain (1950). Thus the pathogen causing white necrotic spots surrounded with water soaked area was identified as *C. cocciniae* Munjal, Lall and Chona.

The colonies of *Colletotrichum* sp showed good sporulation and pink pigmentation on PDA medium. Setae of the fungus were noticed abundant in the old culture. The other morphological characters of the organisms were found similar to that described by Mordue (1971). Thus the brown necrotic leaf spots with brown margin and yellow halo were confirmed to be produced by *C. gloeosporioides* (Penz.)Sacc.

Alternaria sp produced a brownish black velvety growth on PDA medium. The hyphal and conidial characters are similar to that described by Sharma *et al.* (2000) and there by confirming *A. alternata* (Fr.) Keissler as the leaf spot pathogen producing large brown lesion with zonations and yellow halo.

The three fungal pathogens isolated from the three different locations showed same cultural and morphological characters. The cultural and conidial characters of the organism isolated from fruit spot was found similar to the characters of *C. gloeosporioides* (Penz.)Sacc. thereby confirming it as the pathogen of fruit spot.

Histopathological changes brought about by the different pathogens were also observed in the present study. Cercospora infection caused severe distortion upto mesophyll layer and conidiophores were found initiated from palisade tissues, where as other two pathogens caused damage only on epidermal cells. But, the combined infection of *Cercospora* sp *and Colletotrichum* sp affected the distribution pattern of chloroplast resulting in disintegration of chlorophyll grains. Complete death of cells and necrosis were noticed in advanced stages of all infections.

A study on conidial germination of the leaf spot pathogens at room temperature was conducted to find out the minimum time taken for germination by conidia of each organism. Among the three fungi, *C. cocciniae* started germination at 2 hours after incubation and showed 12 per cent germination. Whereas, *C. gloeosporioides* and *A. alternata* initiated the germ tube formation at three hours after incubation. *C.cocciniae* showed cent per cent germination with in seven hours after incubation where as *C. gloeosporioides* and *A. alternata* had taken nine and ten hours respectively for cent per cent germination. From this study, it was clear that the conidial germination was faster in *C.cocciniae,* as compared with the other two pathogens. In the case of *C. cocciniae* the germ tube developed most frequently from the tip cells, but the intermediate cells also showed germination. Jain (1950) also observed the conidial germination of *Cercospora* mainly from the terminal cells and occasionally from intermediate cells, thus supporting the present findings. In the case of *C. gloeosporioides,* the germ tube produced from both ends of the spores and at its end, brownish black irregular shaped appressorium was noticed. All cells of *A. alternata* initiated the germ tube formation, which later developed into hypha.

Resistance of the host plants against pathogen is an important factor in disease development. A genetically resistant plant can prevent infection by pathogen where as a susceptible plant is infected readily. Use of resistant or immune varieties is a simple and effective method in controlling the disease. It is easy to select resistant varieties by screening genotypes, which can be utilized in future for development of resistant varieties. So, nineteen ivy gourd genotypes grown in vegetable field of Olericulture department, Vellanikkara were screened for host resistance against different leaf spot diseases under field condition. Eight genotypes viz., CG8, CG9, CG10, CG11, CG19, CG23, CG27 and CG31 were found highly resistant to the leaf spot diseases showing the Coefficient of Infection value ranged from 0.4 to 3.6 while, CG3, CG5 and CG20 showed resistant reaction. Two genotypes showed moderately resistant reaction and all the rest were moderately susceptible except CG32, a popular local variety which was found susceptible to disease. Vander Plank (1968) reported that the difference in disease resistance exhibited by the genotypes may be due to different types of interaction between pathogen and the genotypes that affected. Many factors are responsible for the resistant type of reaction shown by the genotype. This may include insufficient inoculation load, absence of pathogenic races to that genotype, unfavourable environmental condition and nutrient status of the soil in which the crops were cultivated (Yarwood, 1978; Khan, 1989).

Raju and Peter (1995) reported that CG1, CG9, CG23 and CG27 as promising genotypes of ivy gourd according to their fruit length, fruit weight and fruit girth. According to Joseph (1999), CG11 was high yielder and CG23 as best genotype for their high content of carotene, vitamin C and calcium contents. In the present study CG9, CG11, CG23 and CG27 were found highly resistant to the leaf spot diseases. The genetic characters of these genotypes to resist the interaction with the pathogens along with the good characters observed by the above workers can be utilised for the development of new genotypes having disease resistance and good quality parameters.

From the survey conducted in three locations, to study the seasonal influence on the occurrence of different leaf spot diseases, it was observed that leaf spot due to *Cercospora* and *Colletotrichum* infection were more prevalent during rainy season (June-July). But it was also noticed in February which might be due to the rain showers received during that month whereas, *Alternaria* infection was severe during summer months. Thus, this study indicated that, low temperature, high humidity and high rainfall were the favourable climatic factors responsible for leaf spot symptoms caused by *C. cocciniae* and *C. gloeosporioides* on ivy gourd, whereas, comparatively high temperature, moderate relative humidity and rainfall were favourable for *Alternaria* infection. Mordue (1971) reported that low temperature and high humidity favoured *C. lindemuthianum* causing bean anthracnose. *C. orbiculare* causing bottle gourd leaf spot was found to be favoured by high relative humidity combined with rainfall (Gandhi *et al.*, 1997). Kumar (1999) observed maximum infection of *C. lindemuthianum* on cowpea during the south west monsoon season. These observations are in conformity with the present findings.

For effective management of any disease, the most economic way is the use of resistant varieties along with cultural, chemical and biological methods. Studies conducted on screening of genotypes revealed that only eight genotypes are highly resistant to leaf spot diseases. So the control of leaf spot disease using plant protection chemicals could be adopted as an alternative for this, because, the use of chemicals offer comparatively more effective and speedy action in reducing the disease. Many earlier reports revealed successful control of leaf spot disease by fungicidal application, but constant use of these chemicals gave a chance to develop resistant races of pathogen and cause phytotoxicity and environmental hazards. As an alternative, use of plant extracts and antagonists are followed. Many reports are there showing their effectiveness in controlling different pathogens. So the present study was conducted to select certain plant extracts and antagonists along with some fungicides, which are reported to be effective against the pathogens.

Initially, an *in vitro* evaluation study was conducted to select the best among the fungicides and botanicals against each pathogen. Fungicides such as copper oxychloride, copper hydroxide, captan and mancozeb, each at 0.1, 0.2 and 0.3 per cent concentrations were evaluated. Botanicals used were the leaf extract of Lantana and Adathoda, bulb extract of garlic each at 5,10 and 15 per cent concentrations and neem oil at 0.1, 0.2 and 0.3 per cent concentrations. The *in vitro* evaluation of these fungicides and botanicals against *C. cocciniae* revealed that all the treatments were significantly superior to control, but all of them were not equally effective in inhibiting the fungal growth. Among the treatments, bulb extract of garlic at all the three concentrations recorded maximum inhibition of the pathogen (91.11 per cent). It was followed by captan and copper hydroxide at 0.2 and 0.3 per cent recording a range of 80.99 to 85.32 per cent inhibition over control. Among the fungicides, mancozeb, at 0.1 per cent concentration recorded the least inhibition (53.32 per cent) on the growth of the pathogen. Except garlic, all other botanicals tested, recorded below 50 per cent inhibition on the growth over control. The fungitoxic effect of garlic extract against different plant pathogens have been reported by several workers (Lakshmanan, 1990; Qvarnstrom, 1992; Barros *et al.*, 1995; Kurucheve and Padmavathi, 1997 and Singh and Majumdar,2001). But the search on literature did not provide any information on the effect of garlic on *Cercospora cocciniae*.

In vitro evaluation of fungicides and botanicals against *Colletotrichum gloeosporioides* showed that all treatments were significantly superior to control. Garlic bulb extract at 10 per cent concentration recorded the maximum inhibition and was equally effective with all the three concentrations of copper hydroxide recording 85.66 to 90.0 per cent inhibition over control and also affected complete sporulation of the pathogen. Except garlic bulb extract at all the three concentrations, all other botanicals recorded below 50 per cent inhibition on the growth. So, from the study, it was found that all the three concentrations of copper hydroxide and copper oxychloride and garlic 10 per cent are equally effective in inhibitng the growth of *C.gloeosporioides*. Least inhibition on the pathogen (14.77 per cent) was observed with mancozeb, 0.1 per cent. Madaan and Grover (1980) reported that Difolatan (captafol) or Blitox (copper oxychloride) had shown better effect in controlling anthracnose of bottlegourd caused by *C.lagenarium*. Shivpuri *et al.* (1997) reported anti fungal effects of extracts of *Azadiracta indica, Ocimum sanctum, Allium cepa and Allium sativum* against many fungal pathogens.

In case of *A. alternata,* copper hydroxide 0.2 per cent gave maximum inhibition of the pathogen followed by copper oxychloride and captan, 0.2 per cent. Among the botanicals, bulb extract of garlic at 10 per cent was found most inhibitory, recording 81.55 per cent inhibition over control, which was equally effective as copper hydroxide. Here also, except garlic, all the other botanicals recorded below 50 per cent inhibition over control. No sporulation was observed in all concentrations of copper oxychloride, copper hydroxide and garlic. Among all the treatments, the best effect was recorded by copper hydroxide (at all concentrations) followed by garlic at 10 per cent concentration. Misra and Singh (1971) reported the inhibitory effect of Bordaeux mixture, Blitox-50 and Fytolan against *Alternaria tenuis* and *Helminthosporium oryzae*. Barros *et al.*(1995) reported the inhibition of *A. alternata* and *A. longipes* with bulb extracts of garlic. Singh and Majumdar (2001) also reported the effect of garlic extract in controlling growth of *A. alternata* in pome granate.

Summing up the bioassay results already observed for the three leaf spot pathogens, it was found that among the botanicals, bulb extract of garlic had efficient fungitoxic activity and was equally effective to the fungicides in inhibiting the growth of the three pathogens. All other botanicals viz., leaf extract of Lantana, Adathoda and neemoil were less effective to all the three pathogens. Among the four fungicides tested, 0.2 per cent captan and copper hydroxide were effective against *C. cocciniae* and all concentrations of copper hydroxide and copper oxychloride were found effective against *C. gloeosporioides* and *A. alternata.*

In vitro evaluation of four different fungal antagonists against the three pathogens showed antagonistic effect on the pathogens. *T. viride and T. harzianum* were found more effective due to their faster growth rate and showed complete over growth on the pathogen with in six days. *A. niger* also showed overgrowth on the three pathogens, but not as fast as *Trichoderma* spp., and completed its overgrowth in seven days. *C. globosum* showed a homogenous mechanism against *C. cocciniae* which grow and freely intermingled with the hyphae of the pathogen, whereas with *C. gloeosporioides* and *A. alternata* it showed a slight aversion at first and then showed overgrowth. Among the four antagonists, cent per cent inhibition on growth of all the three pathogens was shown by *T. viride* and *T.harzianum*. They also recorded the highest AI value of 1500 against all the pathogens. Based on the above observation, *T.viride* and *T. harzianum* were selected as the best fungal antagonist against *C.cocciniae*, *C. gloeosporioides* and *A.alternata* and were used for further studies. Antagonistic activity of *Trichoderma species* against different pathogens were well known. *Trichoderma* spp. are mainly used for the disease management of soil borne pathogens, but its efficiency against the aerial pathogens is also reported by many workers (Sychev and Shaprshnik, 1982; Menon, 1996; Bankole and Adebanjo, 1996; Smitha, 2000). Michereff *et al.* (1993) reported the inhibitory effect of *T. viride* and *T. harzianum* on *Colletotrichum graminicola* infecting sorghum leaves.

Three different methods of inoculation were applied for the evaluation of antagonistic property of *P. fluorescens* against the leaf spot pathogens. Ring method of inoculation was standardized and was found to be the best and easy method for inoculation of bacteria around the fungal pathogen. Inhibition zone was not seen against *C.cocciniae*, but 67.3 per cent inhibition of growth was noticed. Inhibition zone of 1 mm and 2 mm was noticed in case of *C. gloeosporioides* and *A.alternata*. with an inhibition of 56.6 and 56.9 per cent respectively over control. Thus, from the *in vitro* evaluation, it was concluded that *P. fluorescens* was also an efficient antagonist to the three pathogens which recorded more than 50 per cent reduction on the growth of the pathogens. So the bacterial antagonist was also selected and used in the succeeding experiments. The antagonistic property of *P. fluorescens* was reported against various pathogens. Srinivasan and Gunasekaran (1998) reported that *P. fluorescens* was effectively used for the management of leaf spot diseases of coconut palm, caused by *C. gloeosporioides*. Sakthivel *et al.* (1986) reported that *P. fluorescens* was antagonistic to *Rhizoctonia solani* in rice under *in vitro* conditions.

During the present study copper fungicides were found to be the effective chemical against leaf spot pathogens under laboratory condition. As the earlier reports (Nene and Thapliyal, 1997) indicated the phytotoxicity of copper fungicides on various crops especially on cucurbits which depends upon the rate and number of applications. So an attempt was taken in order to study the phytotoxic effect of these fungicides on ivy gourd. From the experiment it was found that copper oxychloride(0.1,0.2 and 0.3) and copper hydroxide(0.1 and 0.2) did not show any phytotoxicity on ivy gourd. However, copper hydroxide 0.3 per cent showed marginal yellowing followed by necrosis and complete drying up of middle and young leaves and older leaves were slightly chlorotic. It is worth to mention that copper hydroxide 0.3 per cent showed the above phytotoxic symptoms only after the third spray. Hence, this finding is in conformity with the earlier report stating that the phytotoxicity of copper fungicides on the host is based on the concentration and number of applications of these chemicals.

The next part of investigation was to find out the effect of different fungicides, botanicals and antagonists against leaf spot pathogens under pot culture conditions. In the pot culture studies, all treatments were found superior to control. Among the treatments, 0.2 per cent a copper oxychloride, 0.4 per cent *T. viride*, 10 per cent garlic and 0.2 per cent copper hydroxide were found most effective against leaf spot pathogens recording the lowest severity of 8.0, 8.3, 9.0 and 9.3 per cent respectively. However, spraying of copper oxychloride showed the maximum reduction of 75 per cent over control. In this case, even though a slight increase of disease was noticed 10 days after first spray, a gradual reduction of disease was noticed in the subsequent sprays recording the lowest disease severity of 8 per cent on the 14^{th} day of last spray. Likewise, application of copper hydroxide 0.2 per cent, also showed a drastic reduction in infection after each spray, showing its effectiveness against different leaf spot pathogens. The effectiveness of copper fungicides in controlling leaf spot diseases of various crops were reported by many workers. (Madaan and Grover, 1980; Bharadwaj, 1991; Tahir *et al.*, 1991).

In the present study, it is also observed that eventhough captan showed good inhibitory effect against *C. cocciniae*, a drop in inhibition of pathogen was observed when the study was gradually shifted from lab set up to natural condition. The poor performance of the fungicide in natural condition can be attributed to many reasons including climatic factors, which is beyond the scope of the present study to go deeper into this aspect.

Among the different fungicides used in pot culture experiment, mancozeb fungicide at 0.2 per cent showed the maximum disease severity of 29 per cent and the same trend was also noticed in the lab condition, indicating the ineffectiveness of this fungicide against leaf spot pathogens. Likewise, Yadav and Narain (1993) also reported the least effectiveness of mancozeb against *A. alternata* pathogen on chickpea.

T.viride was found most effective among the different antagonists used against leaf spot diseases. This fungal antagonist has been proved to be a potential antagonist in the control of many vegetable disease pathogens by various workers (Sychev and Shaprshnik, 1982; Smitha, 2000).

Among the botanicals, bulb extract of garlic at 10 per cent recorded highest inhibition of leaf spot diseases. Under *in vitro* condition also this botanical showed the same trend indicating its inhibitory effect on the leaf spot pathogens. Similar results on the inhibitory effect of garlic on vegetable diseases were reported by Mohan and Ramakrishnan (1991) and Barros *et al.*(1995).

In the field evaluation of different fungicides, botanicals and antagonists against different leaf spot pathogens of ivy gourd, all the treatments were found superior to control. Among the treatments, 10 per cent garlic, 0.2 per cent copper oxychloride, 0.4 per cent *T. viride* and 0.2 per cent copper hydroxide were found most effective in reducing the severity of leaf spot diseases. As in the pot culture experiment, here also bulb extract of garlic recorded the maximum reduction of 67.1 per cent over control. Treatment with garlic extract showed gradual reduction in disease severity from the first spraying onwards and recording the lowest disease severity of 9.4 per cent on 14th day of last spraying.The fungicidal properties of garlic against leaf spot pathogens of various crops have been reported by several workers: Barros *et al.* (1995) and Singh and Majumdar (2001) reported the inhibitory effect of garlic extract against *A. alternata*. The anti fungal activity of extracts of *A. indica, A. cepa* and *A. sativum* against different fungal pathogens have been reported by Ram (1997) and Shivpuri *et al.* (1997). Thus the present findings are in conformity with the above results.

Among the fungicides, copper oxychloride recorded the maximum reduction in disease severity of 66.8 per cent, which was followed by copper hydroxide at 0.2 per cent concentration recording 65 per cent reduction over to control. Both copper fungicides were found effective in controlling the leaf spot diseases and showed gradual reduction in disease severity after each spray. Madaan and Grover (1980), Suhag *et al.* (1983) also reported the effectiveness of copper oxychloride against leaf spot diseases of cucurbits caused by *Colletotrichum lagenarium* and *Cercospora sp.* Thus, the present findings are in conformity with the above results. However, a search on literature on the effectiveness of copper hydroxide against leaf spot pathogen of cucurbits revealed no information. So the effect of copper hydroxide on leaf spot disease of cucurbits have been studied for the first time. The per cent disease incidence and severity of fruit infection were also evaluated during the experiment. The same treatments which were found effective against leaf spot diseases also recorded maximum inhibition of disease on fruits. The treatments, T_1 (copper hydroxide),T_2 (copper oxychloride), T_4(garlic) and T_5 (*T.viride*) made the fruits absolutely free from diseases. Among them 0.2 per cent copper oxychloride and 10 per cent garlic were found more effective because from the second spray onwards, the fruits escaped from diseases completely. However, the yield recorded by the various treatments did not show any difference in statistical analysis, but were found superior than the control. During the present study, a bacterial leaf spot infection was also observed on the fruits, so our finding on this effectiveness of copper hydroxide which is having a bactericidal property will be very much useful in controlling both fungal and bacterial infections.

It is also noticed that *T. viride* as the best antagonist in reducing the disease severity which recorded 66 per cent reduction in disease severity over control. There are a number of ways by which antagonists suppress the growth of pathogen and control the disease (Cook, 1990) of which production of antibiotics, volatile compounds, lytic enzymes, induction of host resistance and antagonistic proteins are important. Eventhough, various *Trichoderma* spp. have been reported to be a potential tool for controlling various plant pathogenic fungi in vegetables, the information on its efficiency on leaf spot pathogens of cucurbits are rather lacking.

However, the effectiveness against *C. truncatum* causing brown blotch in cowpea has been reported by Bankole and Adebanjo (1996) supporting this result.

Recalling back the results observed in the present study so far, it is evident that the fungicides 0.2 per cent copper oxychloride and copper hydroxide, 10 per cent bulb extract of garlic and 0.4 per cent *T.viride* are the best treatments for the management of various leaf and fruit spot diseases of ivy gourd.

Summing up the discussions so far, it may be concluded that the present investigation have enriched our knowledge in various aspects of leaf spot diseases of ivy gourd in Kerala, especially the etiology, symptomatology, host resistance, seasonal influence and disease management aspects.

Summary

6. SUMMARY

The present investigation on "Etiology of leaf spot diseases of ivy gourd (*Coccinia grandis* (L.)Voigt)" was carried out to study various aspects particularly etiology, symptomatology, host resistance, seasonal influence and management of diseases.

A survey was conducted at three different locations *viz.*, vegetable field of Olericulture Department, Vellanikkara, and farmer's field at Cherumkuzhi and Elanad, in Trichur district. Three different types of leaf spots and one type of fruit spot were noticed and isolation of pathogens from the infected leaves and fruits showed the association of three different fungal pathogens on leaves and one fungal and one bacterial pathogen on fruits. The pathogenicity of these organisms was proved by artificial inoculation under *in vitro* and *in vivo* conditions. Based on morphological characters, the fungi were identified as *Cercospora* sp., *Colletotrichum* sp. and *Alternaraia* sp. Fungal pathogen associated with fruit spot was identified as *Colletotrichum* sp. Identification and characterization of bacterial pathogen was not conducted. Further studies related to fungal pathogens were carried out.

Studies on symptomatology of various leaf and fruit spots collected from the three locations showed similarities in symptoms produced by the respective pathogens. The leaf spot caused by *Cercospora* sp. were seen on middle aged leaves and appeared as white necrotic distinct spots of 1-2mm diameter with characteristic water soaked area around the spots. The fungus produced conidiophores and conidia in the centre of spots as black dots. *Colletotrichum* sp. attacked middle aged leaves and produced reddish brown spots surrounded by yellow halo of 1-2.5mm diameter. Acervuli seen as black dots in the centre and occassionally shot hole symptoms present. *Alternaria* sp. infected older leaves, produced brown necrotic lesions of 3-5mm diameter with characteristic concentric zonations and yellow halo and infection usually started from leaf margins.

Cultural and morphological characters of the three pathogens were studied in detail. Colonies of *Cercospora* sp. on Carrot Leaf Extract Agar medium showed brownish black coloured mycelial growth with a pinkish tinge. Fusion between vegetative hyphae (anastomosis) was observed in the culture. Measurements of conidiophores from host tissues showed 46.7-155.6µm×3.9µm with 3-6 septa. Conidia measured 23.3-93.4µm×3.9µm with 4-12 septa. Based on the characters, organism was identified as *Cercospora cocciniae* Munjal, Lall and Chona.

Collectotrichum sp, on Potato Dextrose Agar medium showed pinkish white fast growing colonies which turned to brown with pink pigmentation. Acervuli were present. Conidia measured 11.7-15.6µm×3.9µm, hyaline aseptate with oil globules. The organism was identified as *Colletotrichum gloeosporioides* (Penz.) Sacc.

Alternaria sp. on Potato Dextrose Agar medium showed dark brownish black colonies having a velvetty appearance. Conidia formed in chain or free and measured 19.5-31.2µm×7.8-15.6µm with both longitudinal and transverse septa. Based on all characters the organism was identified as *Alternaria alternata* Fr. Keissler.

Germination of spores of *C. cocciniae* started at two hours after incubation from all the cells but more frequently from the tip cells. *C. gloeosporioides* germinated after three hours after incubation from both ends of spores and produced germtubes with appressorium at its end. Spores of *A. alternata* showed germination three hours after incubation from all cells produced germtube which developed in to hypha.

The exotoxin and endotoxin extracted from culture of *C. cocciniae* when spotted separately on leaves of ivy gourd produced yellowing of leaves with water soaked areas on five days after incubation. The endotoxin of *C. gloeosporioides* also showed the same symptoms six days after incubation where as the endotoxin of *A. alternata* produced a slight yellowing of leaves on seven days after incubation. The exotoxin of *C. gloeosporioides* and *A. alternata* had no effect on disease development.

Among the nineteen different genotypes of ivy gourd screened, none of them were found totally free from disease. Eight genotypes showed highly resistant reaction, four were resistant and two were moderately resistant. Four genotypes showed moderately susceptible reaction and the local variety showed susceptible type of reaction. Out of the eight highly resistant genotypes, CG9, CG10, CG23 and CG 27 showing CI value less than 1.0 were found to be the best varieties considering their resistance to leaf spot diseases.

Studies on seasonal influence on the incidence and severity of leaf spot diseases revealed the effect of temperature, relative humidity and rainfall on disease development. For *Cercospora* and *Colletotrichum* leaf spots, low temperature, high humidity and high rainfall seemed to favour the disease, so they are prevalent in rainy season. *Alternaria* infection was noticed during summer periods favouring high temperature, moderate relative humidity and rainfall.

For the management of leaf spot diseases, an *in vitro* evaluation was conducted on the effectiveness of fungicides, botanicals and antagonists against leaf spot pathogens. Based on the result of the *in vitro* evaluation, 0.2 per cent copper oxychloride, copper hydroxide, captan and mancozeb, 10 per cent leaf extract of Lantana, Adathoda, bulb extract of garlic and 0.2 per cent neem oil were selected. The fungal antagonists *T. viride*, *T. harzianum* and the bacterial antagonist, *P. fluorescens* were also selected for pot culture studies.

During the pot culture experiment, the fungicides copper oxychloride, copper hydroxide (both at 0.2 per cent), the botanicals garlic (10 per cent) and neem oil (0.2 per cent) and the antagonists *T. viride* (0.4 per cent) and *P. flourescens* (0.2 per cent) were found effective in reducing the disease severity and were selected for the field evaluation.

Field experiment on the effectiveness of selected fungicides, botanicals and antagonists against leaf spot diseases confirmed the efficiency of 0.2 per cent copper oxychloride, copper hydroxide, 10 per cent garlic and 0.4 per cent *T. viride* for the management of various leaf spot diseases of ivy gourd under natural condition. The treatments also showed their effectiveness on controlling the fruit infection. However, no significant difference was observed on the yield recorded by the application of various treatments except control.

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* Originals not seen

Appendix

APPENDIX-I

MEDIA COMPOSITION

1. POTATO DEXTROSE AGAR

Potato	: 200 g
Dextrose	: 20.0 g
Agar	: 20.0 g
Distilled water	: 1000 ml

2. CARROT LEAF EXTRACT AGAR

Carrot leaf	: 300 g
Agar	: 20.0 g
Distilled water	: 1000 ml

3. COCCINIA LEAF EXTRACT AGAR

Coccinia leaf	: 300 g
Agar	: 20.0 g
Distilled water	: 1000 ml

"ETIOLOGY OF LEAF SPOT DISEASES OF IVY GOURD (*Coccinia grandis* (L.) Voigt) AND THEIR MANAGEMENT"

By

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ABSTRACT OF THE THESIS

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ABSTRACT

A study on various aspects of leaf spot disease of ivy gourd was conducted at College of Horticulture, Vellanikkara during 2002-2003. Etiological studies revealed that *Cercospora cocciniae* Munjal, Lall and Chona, *Colletotrichum gloeosporioides* (Penz.)Sacc, and *Alternaria alternata* Fr.Keissler were the main pathogens causing different leaf spot diseases of ivy gourd. *Colletotrichum gloeosporioides* was found as the main pathogen causing spots on fruits of ivy gourd. Among the nineteen ivy gourd genotypes tested eight genotypes were found highly resistant to different leaf spot diseases. For *Cercospora* and *Colletotrichum* leaf spots, low temperature, high humidity and high rainfall were found to favour infection, so they were prevalent in rainy season. *Alternaria* infection was noticed during summer months favouring high temperature, moderate relative humidity and rainfall.

From the *in vitro* evaluation conducted for testing the efficacy of fungicides and botanicals, 0.2 per cent copper oxychloride, copper hydroxide, captan and mancozeb, 10 per cent leaf extract of Lantana and Adathoda, bulb extract of garlic and 0.2 per cent neem oil were selected. The fungal antagonists *Trichoderma viride*, *T. harzianum* and the bacterial antagonist, *Pseudomonas fluorescens* were also selected for pot culture studies. During the pot culture experiment, copper oxychloride, copper hydroxide(both at 0.2 per cent), garlic (10 per cent) and neem oil (0.2 per cent) and the antagonists, *Trichoderma viride* (0.4 per cent) and *P. fluorescens* (0.2 per cent) were found effective against leaf spot diseases and selected for field evaluation. From disease management study conducted in the field, 0.2 per cent copper oxychloride, copper hydroxide, 10 per cent garlic and 0.4 per cent *Trichoderma viride* were found equally effective in controlling both leaf and fruit infection in ivy gourd.